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- (54) **ISOLATED NUCLEIC ACID MOLECULES FROM THE GENOME OF CITRUS SUDDEN DEATH VIRUS AND USES THEREOF**
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- (51) **Int. Cl.**  
**C12N 15/82** (2006.01)  
**C12N 15/87** (2006.01)  
**A01H 1/00** (2006.01)  
**C12N 7/00** (2006.01)  
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**CI2Q 1/70** (2006.01)

- (52) **U.S. Cl.**  
CPC ..... **C12N 7/00** (2013.01); **C07K 14/005** (2013.01); **C12N 15/8283** (2013.01); **CI2Q 1/701** (2013.01); **C12N 2770/40022** (2013.01); **C12N 2770/40051** (2013.01)

- (58) **Field of Classification Search**  
CPC ..... C12N 7/00; C12N 15/8283; C12N 2770/4022; C12N 2770/40051; C07K 14/005; C12Q 1/701  
See application file for complete search history.

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*Primary Examiner* — Li Zheng(74) *Attorney, Agent, or Firm* — Lowe Hauptman & Ham, LLP(57) **ABSTRACT**

The present disclosure relates to nucleic acid molecules comprising the genome of the Citrus Sudden Death Virus (CSDV), the causative agent of Citrus Sudden Death (CSD) disease. The cloned CSDV nucleic acid molecules can be used as probes or can be used to design oligonucleotide primers useful in assays, such as a polymerase chain reaction, for detecting the presence of CSDV in biological samples, particularly leaves, roots and other tissues or organs of plants, such as plants from the genera *Citrus* and *Poncirus*. The disclosure also comprises the nucleic CSDV nucleic acid molecules, in whole or part, as well as transgenic plants, such as monocots and dicots, containing the CSDV nucleic acid molecules, in any kind of combination, so that expression increases resistance to CSD disease.

**8 Claims, 19 Drawing Sheets**

## Figure 1

### Comparison of genome sequence of CSDV with the genome sequence of the Oat Blue Dwarf Virus (OBDV)

OBDV						
CSDV		-----GTCCCCCTGTGATCGTCTCCCGCCCTCCAGCCGGAAAGATA	TTTTGCTTAAC	TTTC		
	1				60	
OBDV						
CSDV		-----TTTGCACTCTTACGCTCAGATCTACGTGCC	TTAGGT	CATCTAAGCCG	CATGGATCGCAT	
	61				120	
OBDV						
CSDV		-----CTCTGCCCGATTCCC	GCGCCG	CTTCCGCCGGCCGACC	GAGTACACTCC	CATACCC
	121				180	
OBDV						
CSDV		-----GTGTCCAAG--GTGTCA	TATTAT	-TCCGCTCAG	TTTCAGA	GTCTGCCG
	181				240	
OBDV						
CSDV		ACACACTCACCCACTCCTAC	CCCCGAGGTG	TCTCACCTCCGG	GGCCATTTCAC	ACACCCTGTCT
	241				300	
OBDV						
CSDV		GAATTCTCCAAG-CATCCCGCCAAAAGCCGGCTGCTTAA	ATCTG	ATCTTGATCTTCTCC	ATCT	CACCT
	301				360	
OBDV						
CSDV		TGTC	CAAGTGTC	--GTTATGACCACATACG	CGCTT	CCCCACCCGACC
	361				420	
OBDV						
CSDV		TCCTTCGCACTATCA	CTGGGGGTG	TTGAAGGATGTTATCGA	AAACCC	CTCTCGTCCACC
	421				480	
OBDV						
CSDV		ATCCACAGAGACACGATCGCAGC	ACCCCTCATGGAGAC	CCCTCGCCTCG	CCCTAACCG	AGAC
	481				540	
OBDV						
CSDV		TCCCTTCGGA	CTTCCCTGGGCC	GCTCCCGC	CTCCGCC	CTCCAGGAATGT
	541				600	
OBDV						
CSDV		GGCATCACCGTCGCCGCCACCG	GGTTCAAAGCTCAT	CCCCACCC	GTGTC	ACAAAAACCATC
	601				660	
OBDV						
CSDV		GAGACCA	CACCTCTCCACA	AGGTTGGC	CTCA	ATGCCCAGTC
	661				720	
OBDV						
CSDV		TTCATGAAGCC	CTCGCAAGTTC	CGCAA	CTCCAG	CCAACTTCTCCGCACTC
	721				780	

Figure 1 (continued)

Figure 1 (continued)

OBDV	CTCATCTTCCGGCCCCCTCCACCCCC--AAGCTTCGCTTTCTCCAGCGCAACT-	
CSDV	CTGATTGACCTCCCATTCTCCGACCCTTAGCTGTCCCTTCCACATGCGCCTAACTG	
	* *	
	1681	1740
OBDV	-----CCACCGGGCCGGTCTTCTCCGTGGCTCC---GCCTCGAG-----	
CSDV	GGCGCCAAGCACCCCTGCTCTTGCACATTGCTTCCCTGCTGCTCCCAGGCCACATGGCCC	
	* *	
	1741	1800
OBDV	TTTGAGGCTTCCCTCTCGCC---CCACAACTCGCCCGTCGCTTCC---ATTC	
CSDV	CTAAAGGTTGGCCCTCGCACTCGCTGCTGCTCCAGTCTGCCTGTCTTGCGGAAATTC	
	* *	
	1801	1860
OBDV	CTCGCTCGCTTCTCCCCAGAA-----ACCCATCGACCCCTGGGTCGTC---G	
CSDV	ATTGGTCCCGATTCTCCACAGGACATGCATGACAGCTATCATGCCATGTTCATCCACAG	
	* *	
	1861	1920
OBDV	C---GAGCTCGCTGTCG-CGTTGCTATAACCGCCGCCCTCC---CTCGCCGTTCGCTG	
CSDV	CTTGGGGCCTACTCTCACCAAGGCTATGCTGCGATAGGGCCCCCTTCTGCC	
	* *	
	1921	1980
OBDV	GTTCTT--CGGCCCCGACAC---CCCCAAGCCATGC---ACGACCGATACCACACCATG-	
CSDV	ATCCCTGTTCCAGCTCTGACTTAAGGGTGTGGAGCTCAAGTTCTGGAGAAGGACTACCTCCGAG	
	* *	
	1981	2040
OBDV	--TTCCACCCC---AGAGACTGGCCCTCACCTGCC---AGGGGCCCCATCTCAT	
CSDV	ACTTCCATCCATTAAAGGTGTGGAGCTCAAGTTCTGGAGAAGGACTACCTCCGAG	
	* *	
	2041	2100
OBDV	GTCG-----CGCTCAGCTTC---TCCCCCTTCCCC---ACCCACCTTCGCC	
CSDV	TCCGCTTCATCAACCGGGGGCATGGACTCCGTCGIGGGCGCAACCGCTTCATCA	
	* *	
	2101	2160
OBDV	ACT--CCCGCTCCCGACTCCCGACG-----TGAACCCCTCCAGCCACCC-----	
CSDV	ACTGGTCCGGATCGGCCAACCGAACACGAGTGCTGCCCCCCCACCTCCCATCGAATCC	
	* *	
	2161	2220
OBDV	--TCCGCTCCACCC-----TCGACCCACGAGC-----	
CSDV	AAAGTTACCTTGCCCAACCCATTGAGAGTGTGGCACCTGTAGTTCCAGGAGCAGGAGAA	
	* *	
	2221	2280
OBDV	--CGGCTCC-----CGCCGATCTC-----GAGCCCCAAGCT	
CSDV	CCTCCGCAGTCGGCTTCATCAACCGGGGGCATGGCTCCGCGTGACCCGCAARGTG	
	* *	
	2281	2340
OBDV	CCTCCGGC--CCACGCC-----CCCCAGACCGAGCCTCCGAGT--CCCGTG-----	
CSDV	GCTTCATCAACCACTCCGGATGCTCCACCCCTGGAGCTCAGCGTGACCCCTCCAAAGACT	
	* *	
	2341	2400
OBDV	ATCGAGC--AAGAAGCGCTCCGAATCCCCCTC---CCGCTCTGCCGCTT-----	
CSDV	ATCTATCCTATTGACCACTCCAGAACGACTTCGGCCCTTGCCGTTGCTCCGCTGTGAA	
	* *	
	2401	2460
OBDV	-----TCTGCTCCACCCCTCCGCTTC-CGCGCCTCACTTGCCCCAAC--ACCC	
CSDV	CCACTTCAGCTGGCCCCGTCCTCCACTCTCACCGCTCGGATCATAAAAGAAGCC	
	* *	
	2461	2520
OBDV	TCGGGCCCCCGAGCCTCCCTCGCCGACG-----GCTTCCGAGCAGGCCGCGTCCCTCATC	
CSDV	CAGGACGCCAGGCTTTCTCGGCCCTCCAGGCCCTGGGCTCGCTCCACCCACCA	
	* *	
	2521	2580

Figure 1 (continued)

Figure 1 (continued)

Figure 1 (continued)

OBDV	CCCTACTCCCGCTCATTCCAGTCGATCTCGCTTCGCCGCCGTCAAGCCTTCCGAC	
CSDV	CTCCACCCCAGCTC-----	TCAAGCCTGATTTC
	*****	*****
	4321	4380
OBDV	CGGTCAAGACGCTCTCGTGGGCCCTATGCCGCGGTGACGGGAAACCAACGCCCT	
CSDV	CAAGGGGACGTCTATAATCTCAGCACCCATAGTTCTCGGCTCCGGGAGCTCAATGCCCT	
	*****	*****
	4381	4440
OBDV	CGCATTTGACACCTCCTCCGTGCCCAGAACTCGCCGCCCTTCATTTGATCTTCCCTCG	
CSDV	CAAGTCTCCTCTACTTCCCTCCCGAGACTCGCCGCTCCACTGGGACATTCCATCT	
	*****	*****
	4441	4500
OBDV	TTCCGCCCCA-AGCCCCACCGCCTCCCTTGACCCAGCCCTTCTGGGACCGCCTTGA	
CSDV	GCCATCCCTGAGAGTGCACAGACGGGACTCCACTGAGGCCACCACCTCCATCCA-GA	
	*****	*****
	4501	4560
OBDV	GCCC GTT TACCC CGG C GAA ACCTT CG AAA ATT TG GT CG CC CA TT CC TT CG G CT C A CG A	
CSDV	GCC AGT CT ACCC CGGG GAA CT TT GAG A AT CT TG CT G C C A CT TT CT C C C T G C C A CG A	
	*****	*****
	4561	4620
OBDV	CCCCACTGACCGCGAAATCACTGGCTCGCAGCTTCAACCAAGTTCCCCATGCGA	
CSDV	CCCAACCGATCGTGAGATCTACTGGCAGGGTCAGCTGTCACCAAGTCCCACATGGA	
	*****	*****
	4621	4680
OBDV	TAAGGACTACCACCTCGCGCTCAGCCAATGACGCTCTCGCTCCCATCACGACTCCAA	
CSDV	CAAGGAATTCCATTGGCTGCACAACCCATGAGTCTCTGGCTGCCGTCATCAAGAGAA	
	*****	*****
	4681	4740
OBDV	GCACGACCCCAACCTCCTTGCCTCCATCCAGAAACGACTTCGATTTGACCCCTCCG	
CSDV	GCAAGATCCCACCTACTGCCAGCTTCATCCAAAAGAGACTCCGTTCCGCCCTCGA	
	*****	*****
	4741	4800
OBDV	CTCTCCCTACCGAATCTCCCTCGTGACGAGCTGCTGGCCAGCTCCTCTACGAGAGTCT	
CSDV	CAAGCCCTACCAAGATCACCCAAAAGATGAAATCTGGCCAGCTCCTCTTGAAGGCCCT	
	*****	*****
	4801	4860
OBDV	CTGCCGCGGTATCATCGTCCCCAACCAACCACCCACCCCTTCGATGAGGCCCTTTCGT	
CSDV	CTGCCGAGGCTACACAGATCTCATTCACTGAGGCCCTTGATCCCGTGCCTTTCGC	
	*****	*****
	4861	4920
OBDV	CGAGTGTATCGACCTGAACGAAATCGCTCAACTCACCAAGCAAACACTCAGGCCGTCTCAT	
CSDV	CGAGTGCATCAATCTCAATGAGTTGCCAGCTCTGCCAACAGCCAGGCTACTATTAT	
	*****	*****
	4921	4980
OBDV	GGCACACGCCGCCGCTCTGACCCAGACTGGCCTGGTCCCGGTCCGGATCTTCAGCAA	
CSDV	GGCAATGCTGCCGCTCACACCTGATTGGCGTGGAGCGCAGTTCGCATCTTCTCAA	
	*****	*****
	4981	5040
OBDV	AACCCAGACAAGGTCAACGAAGGTTCGATCTTGGACCTGGAAAGCTTGCCAGACCC	
CSDV	GACCCAAACACAAGGTGAATGAAGGGTCAATTTCGCTCTGGAAAGGCCCTGCCAAACTTT	
	*****	*****
	5041	5100
OBDV	CGCTCTCATGCACGACGCCGCTGTTCTGCTCCCTGGCCCGTCAAGAAGTATCACCGCT	
CSDV	GGCTCTCATGCATGATGCTGTTCTAACCTGGGCCCTGTCAAGAAGTACCAAGCGAGT	
	*****	*****
	5101	5160
OBDV	CTTCGATGCTCGAGACCGCCGCCACCTCTACATCCACGCCGCCAGACGCCCTCTTC	
CSDV	CTTTGATCAGAGAGACCGACCCGACACCTTACATCCATGCAAGAACACTCCATCACA	
	***	***
	5161	5220

Figure 1 (continued)

Figure 1 (continued)

**Figure 2**

## **Comparison of CSDV polypeptide domains with those from others Tymoviridae virus (OBDV, GAMaV and GFkV)**

a) Methyltransferase/Protease/Helicase/ RNA-dependent RNA polymerase)

Figure 2 (continued)

OBDV	----SAPEPPSP-----	TASEQQAASLIPAP---	SSALVVEPSGVVSASS
CSDV	EPLQPAPVPSTPLTVSDHKEAQDAEALSSALQALGLAPTPPAPQSQNLTVESSGAMHASS		
	781		840
OBDV	WGATNQPADQVDDSPLARDPSASGPVRFYRDLFPANYAGDSGTDFRARASGRSPTPYPA		
CSDV	WDQLSSPSSWDPSPLARDSSASGPGGMYSOLFPAPYLPGTQFIFRSRANGRANIPYPD		
	841		900
OBDV	MDCLLVATEQATRISREALWDCLTATCPDSFLDPKSIAQHGLSTDHFVILAHRFSILCANF		
CSDV	MDCLLLSIEQATRLPKEARLWDTLCATCPDSLDPDTIRRVGLSTDHFAILAHYSLRCRF		
	901		960
OBDV	HSAEHVIQLGMADATSIFMINTAGSAGLPGHFSLRLGDQPRALNGGLAQDLAVAALRFN		
CSDV	HTAHGVIELGMADATSSFDIDHTAGN---PGHFSLRQSATPR-LNGGIAQDLAVAALRFN		
	961		1020
OBDV	ISGDLLPLTRSHTYRSWPKRAKNLVSNMKNGFDGVMASINPIRPSDAREKIVALDGLLDI		
CSDV	IDGTLPLIRSHTVYSTWPKRAKNLSNMKNGFDGIMANIHPKTNTESREKILALDSQLDI		
	1021		1080
OBDV	ARPRSVRLIHIAGFPGCGKTHPITKLLHTAAFRDFKLAVPTTELSEWEKELMKLSPSQAW		
CSDV	AVRRSVRLIHIAGFPGCGKFPISRLLRTPTFRNFKVAVPTVELRAEWKTITGLPASEAW		
	1081		1140
OBDV	RFGTWESSLLKSARILVIDEIYKLPRGYLDLAIHSODSIEFVIALGDPLOGEYHSTHPPS		
CSDV	RIGTWESSLLKSARVLVIDEIYKMPRGYIDLAIHSOPTIEMVIALGDPLOGEYHSTHPPS		
	1141		1200
OBDV	SNSRLIPEVSHLAPYLDYYCLWSYRVPQDVAAFFQVQSHNPALGFARLSKQFPTTGRVLT		
CSDV	TNSRLLSEPQHLSMYLDFYCLWSHRVPQNVAAFFHVKTTSKQPGFCRYQRELPPNS-RILA		
	1201		1260
OBDV	NSQNSMLMTQCGYSAVTIASSQGSTYSGATHIHLDRNSSLSPNSIVALTRSRTGVFF		
CSDV	NSQNAUGHTLQCGYAAVTIASSQGSTYENAAICHLDRNSSLSPAHSMVALTRSKVGVIF		
	1261		1320
OBDV	SGDPALLNGGPNNSLMFSAFFQGKSRHIRAWFPTLFPTATLLFSPLRQRHNRLLTGALAPA		
CSDV	TGDPQAQLSNAPSSNRMFSEFFSGRTRPLHDWFHNEFPKATVLTEPLKTRGPRLTGAAS--		
	1321		1380
OBDV	QPSHLLPDPLSLPPLPASGPYRSRSFVRSRFAAAVKPSDRSDVLSWAPIAVGDGETNAP		
CSDV	-----PYSKAVPIRQASTPALKPDEQGDVIISAPIVLGSGELNAP		
	1381		1440
OBDV	RIDTSFLPETRRPLHFDLPSFRPQAPPSSDPAPSGTAFEPVYPGETFENLVAHFLPAHD		
CSDV	QVSSHLPETRRPLHWDIPSAPIESATRPDSTEPTTSHEPVYPGETFENLAHHFLPAHD		
	1441		1500
OBDV	PTDREIHWRRLQLSNQFPHVDFKEYHLAAQPMTLLAPIHDSKHDPTLLAASIQKRLRFRPSA		
CSDV	PTDREIYWGQLSNQFPHMDFEFLHAAQPMSSLAAVHQEKQDPPTLLPASIQKRLRFRPSD		
	1501		1560
OBDV	SPYRISPRDELLGQLLYESILCRAYHRSPTTHPFDEALFVECIDLNEFAQLTSKTQAVIM		
CSDV	KPYQITPKDEILGQLLFEGLCRAYHRSPEFHTEAFDPVLFACINLNNEFAQLSSKTQATIM		
	1561		1620
OBDV	GNARRSDPDWRWSAVRIFSKTOHKVNNECGISFGCAWKACQTLALMHDAVVLILGPVKYORV		
CSDV	GNARRSDPDWRWSAVRIFSKTOHKVNNECGISFRSWKACQTLALMHDAVVLILGPVKYORV		
	1621		1680

Figure 2 (continued)

OBDV	FDARDRPAHLYIHAGQTSSMSLWCQTHLTAVKLANDYTAFDQSQHGEAVVLERKKMER	
CSDV	FDQRDRPRHLHYIHAGNTPSQMSNWCCQHLLTAVKLANDYTAFDQSQHGEAVVLERKKMER	
	*****	*****
	1681	1740
OBDV	LSIPDHILSLHVHLKTHVETOFGPLTCMRLTGEPGTYDDNTDYNLAVINLEYAAHVPTM	
CSDV	LSIPQALIDLHHLKTHVSTQFGLTCMRLTGEPGTYDDNSDYNLAUVNCEYMAANTPTM	
	*****	*****
	1741	1800
OBDV	VSGDDSSLDFEPPLRRPEWVAIEPLLALRFKKERGLYATFCGYYASRVCVRSPIALFAKL	
CSDV	VSGDDSSLREPPTRPEWVILQPLLSLRFKKERGRYATFCGYYASHVGCVRSPVALFAKL	
	*****	*****
	1801	1860
OBDV	ATAVDDSSISDKLAAYLMEFAVGHSGLGSALPLSAVPFQOSACFDFFCRRAPRDLKLA	
CSDV	AIAVDDGSISDKMASYLSEFALGHSLGDHLWEALPLEAVPFQOSACFDFFCRRAPRHLKLS	
	*****	*****
	1861	1920
OBDV	LHLGEVPETIIQRL-SHLSWLSHAVYSLLPSRLRLA1HSSRQHRS1LPEDPAVSSLQGEL	
CSDV	LMLGEVPESI1ARIGSSLKWAHAIYTTLSSAARVAILRSSRNSRSMPDDPTTLLQGEL	
	*****	*****
	1921	1980
OBDV	LQTFHAPMPSLPSLPLFGGLSPDNILTPEFRITALYESSAYPTPPNSFTMSGIHASQVG	
CSDV	LQHFQVPMFQSDTLLPLTGSSA1LTPEAFTSLAFSMAS-----DAQAG	
	***	***
	1981	2040
OBDV	PPPASDDRTDRQPSLPLAPRIVESSLAVPHVDVFPQWAVASYAGDSAKFLTDDLSGSSH1	
CSDV	PAPSRRDRVDRQPRPLPAAPRVAEVGLNAPSVDYFPOWVVASYDGSEAKNLSSDLSGSATL	
	***	***
	2041	2100
OBDV	SRLTIGYRHAEELISAEEFAPLAAAFAKPISVTAVWTIASIAPATTTELQYYGGRLLTLG	
CSDV	TKVMANYRHAEELSVELEVCPLAAAFAKPISVSAVWTIASISPASASETSYYGGRLETVG	
	***	***
	2101	2160
OBDV	GPVLMGSVTRIAPADLTRLNPVIKTAVGFTDCPRTYSVYANGGSANTPLITVMVRGVIRL	
CSDV	GPVLMSSTHLPADLTRLNPVLKGPKVYTDCPRTFSYVSNGGTKGTLCTIILRGVVRL	
	*****	*****
	2161	2220
OBDV	SGPSGNVTAT	
CSDV	SGPSGNLLA--	
	*****	***

(b) Coat Protein 1

GAMaV CSDV	SSAPQLTSEAFSLTLAQSMASPNVQAGPPPPSDDRTDRQQPLPRAPIRVEDASAIPFVDY SSAPILTPEAFSTSLAFMAS-DAQAGPAPSRRDRVRDQRPLPAAPRVAEVGLNAPSVDY ***** * .**** : ** **** : .***** .* . ***.***** * * * * . . * * * * 1	60
GAMaV CSDV	PFQWVVASYDGSTAKNLTDVLSGSKTLSTITANYRHAELLSVELEFAPLAGSFSKPITLS PFQWVVASYDGSEAKNLSDLGSATLKVMANYRHAELTSVELEVCPAAAFSKPKISVS ***** * .**** : ** **** : .***** .* . ***.***** * * * * . . * * * * 61	120
GAMaV CSDV	AVWTVGTSITPATTETSYGGGRITIGGPVLMNSTTAVPADLRLRNPIIKDQIISYTDPCR AVWTIASISASPASETSYGGGRIFTVGGPVLMNSTTHLPADLTRLNPVLKGPVKYTDPCR ***** * .**** : ** **** : .***** .* . ***.***** * * * * . . * * * * 121	180
GAMaV CSDV	FSYSVYANGGTAGTNLVTLIRGVVRLRSPSGNLLA FSYSVYNSGGTKGTCNLCTIILRGVVRRLSGPSGNLLA ***** * .**** : ** **** : .***** .* . ***.***** * * * * . . * * * *	

Figure 2 (continued)

## (c) Coat Protein 2

GAMaV	MASPNVQAGPPPSDDRTDRQPLPRAPIRLVEDASAI	P	F	V	D	Y	F	Q	W	V	V	A	S	Y	D	G	S	T	A	K	N	L
CSDV	MAS-DAQAGPAPSRRDRVDRQPLPAAPRVAEVGLNAPSVDYPFQWVVASYDGSEAKNLS																					
	*** : .****,* . ***.**** * * ***; . * . * * * * * * * * * * * * * :																					
	1																					60
GAMaV	DVLSGSKTLSTITANYRHAELLSVELEFAPILAGSFSKPITLSAVWTVGSIITPATTETSY																					
CSDV	DDLGSATLTKVMANYRHAELLSVELEVCPILAAFSKPISVSAWTTIASISPASASETSY																					
	* **** *; : * * * * * * * * . * * * ; * * * * ; * * * * ; * * * * ; * * * * :																					
	61																					120
GAMaV	YGGGRVITIGGPVLMNSTTAVPADLRLRNPIKDQISYTDCPRFSYSVYANGTAGTNLVT																					
CSDV	YGGRLFTVGGPVLMSTS THLPADLTRLNPVLKGPVKYTDCPRFSYSVYSNGGTGKTNLCT																					
	*****; *; * * * * , *** ; * * * * * * ; * . * . * * * * * * * * * * * * * :																					
	121																					180
GAMaV	VLIRGVVRLRSPSGNLLA																					
CSDV	IILRGVVRSLSGPSGNLLA																					
	: ; * * * * * * * * :																					

## (d) Putative Movement Protein

GFKV	MTSRAPSPPTPPCPSPPALKSSPSPVPTATPASPPLKELSNPLPPPPPPTPRPSTSAGPST																					
CSDV	MISLALPLSPKSWPTTDMLSSHLLSWSRALLQP-----SPSPSLCRPSGPLPPSL																					
	* * . . . . : * . * . : * . * . * . * . * . * . * . * . * . * . * . * . * .																					
	1																					60
GFKV	PLPPPALARSSPSSALNASRGAPSTSPPSSSPSSPASTPPSRTPSPTAPASPVASTA																					
CSDV	QLPP--LKPPTMAVDSSLALAVLSSCPAPPISLLISPASILCSRAPSSQTAPDSPTPSTP																					
	*** *; : . . . . : . . . . * . * . * . * . * . * . * . * . * . * . * . * .																					
	61																					120
GFKV	MTPASPSVPPPSAAPSSAALSSAPPSTA PLPRHEPRPPPPLQPPP C VRV PRSV																					
CSDV	MAEPRAPISAPSSSGELS---ASAAPPVIFSLRASSGEKGHLLVSAR-----																					
	* : . . . . . * . : * . * . * . * . * . * . * . : .																					
	121																					180
GFKV	AFPLPLARELPPRLPPAPYLHPLLARLAPLRLRPPDLPSPPLSPPLSPLSPISPLHA																					
CSDV	-----																					
	181																					240
GFKV	PAPPKHDPVILLPALSIAISRRAPDLLRLLSLLSPPSLFLFLFTLLSIHFSPFFI FILLSL																					
CSDV	-----																					
	241																					300
GFKV	LLLLQFPRT																					
CSDV	-----																					

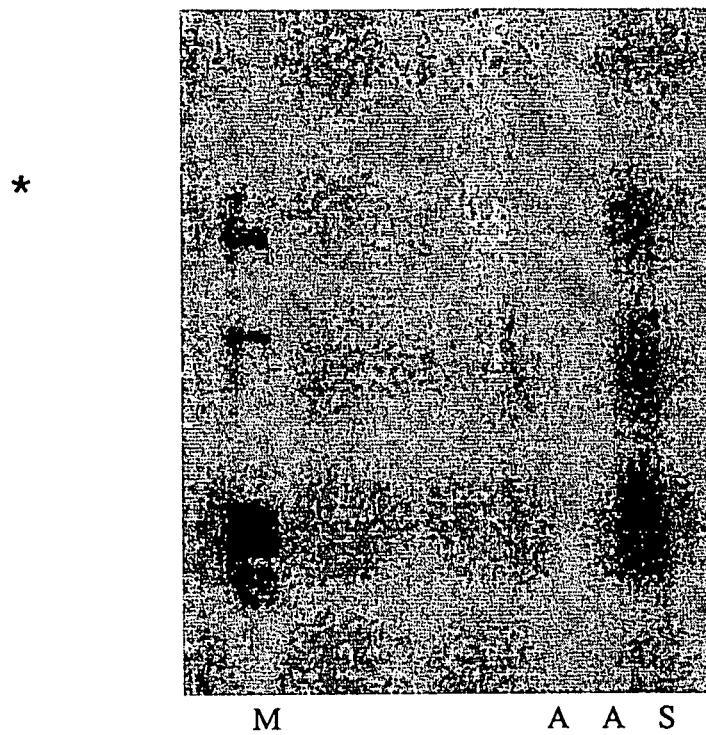
**Figure 3**

**Agarose gel electrophoresis of DNA fragments amplified with primers  
based on the CSDV nucleic acid sequences**



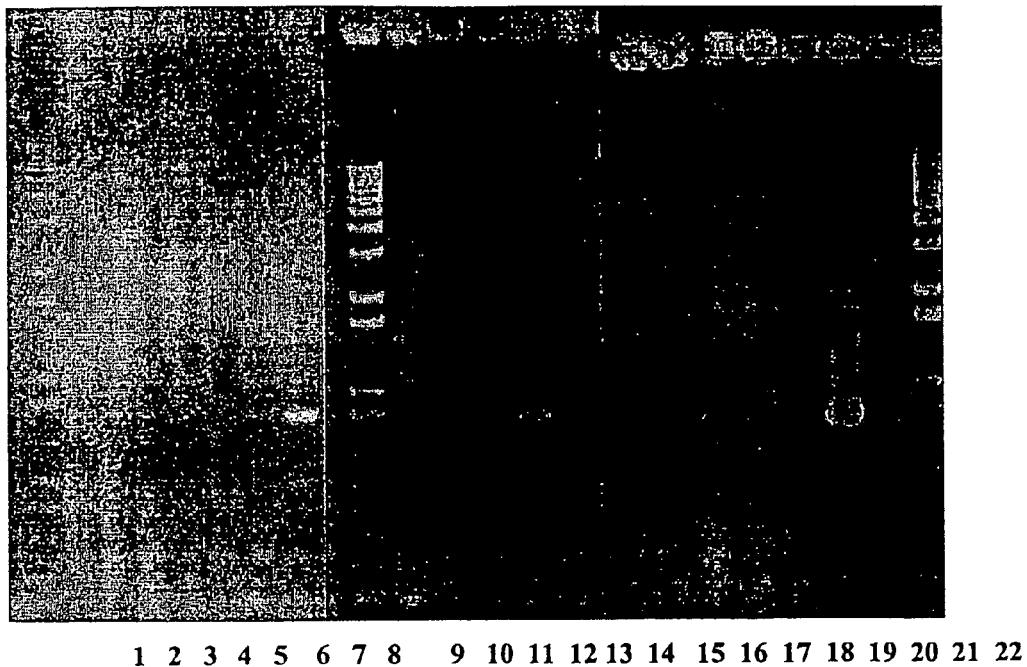
**Figure 4**

**RNA gel blot analysis of RNA samples from CSD-symptomatic and -  
asymptomatic plants**



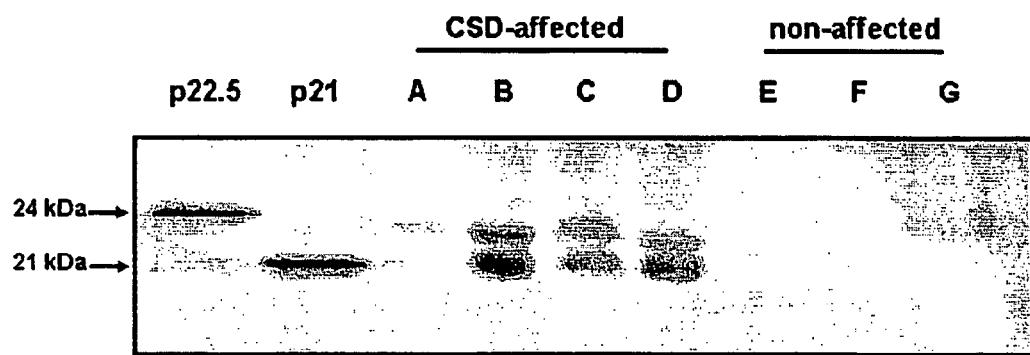
**Figure 5**

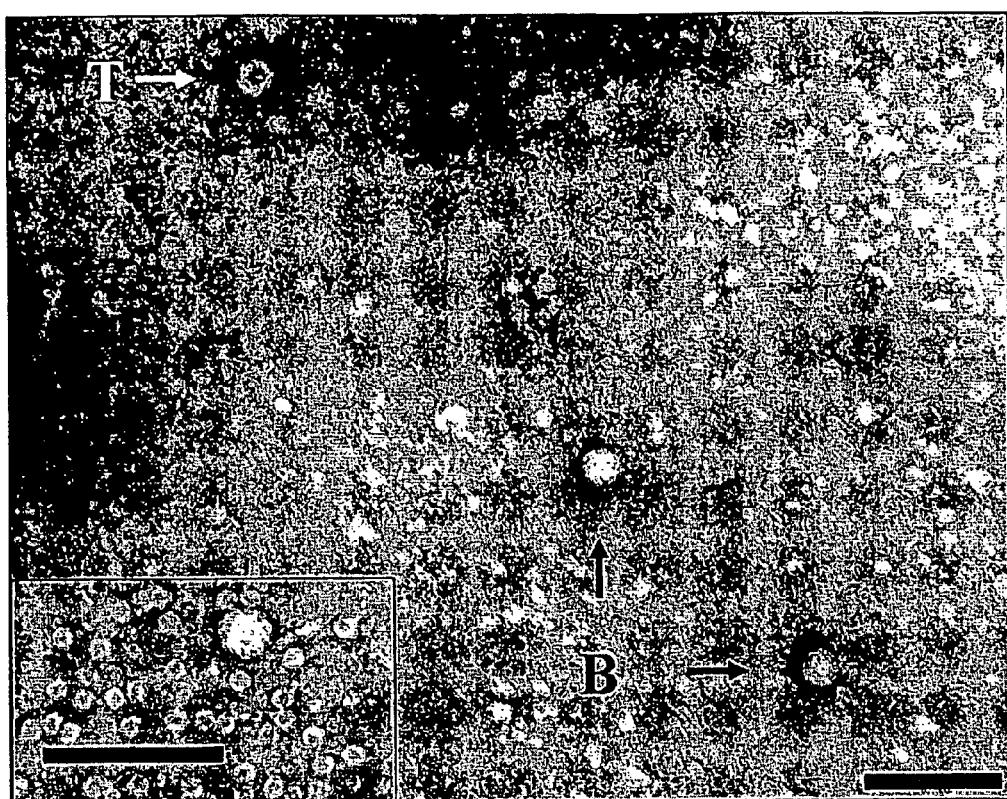
**Agarose gel electrophoresis of PCR products of a DNA fragment amplified  
with primers based on the CSDV nucleic acid sequences**



**Figure 6**

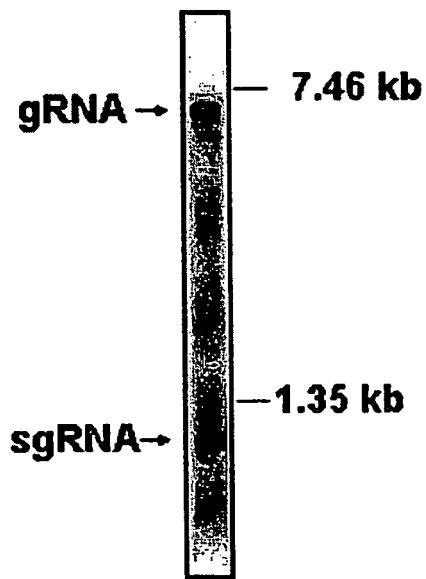
Western blot detection of the CSDV Coat Proteins in the crude protein extracts of CSD-affected and unaffected citrus trees

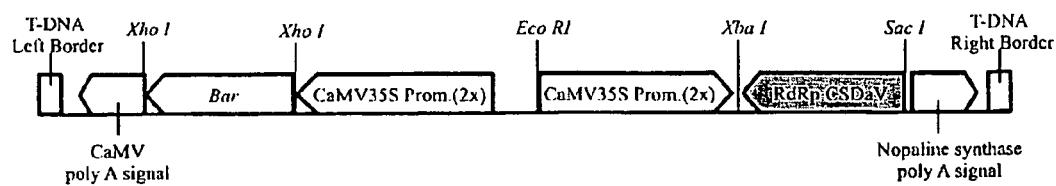


**Figure 7****Electron microscopy of CSDV purified from infected citrus tissues**

**Figure 8**

**Northern blot hybridization of total nucleic acids extracted from purified  
CSDV preparation**



**Figure 9****Drawing of TDNA insert fragment correspondent to pTYMO-AS vector**

## 1

**ISOLATED NUCLEIC ACID MOLECULES  
FROM THE GENOME OF CITRUS SUDDEN  
DEATH VIRUS AND USES THEREOF**

RELATED APPLICATIONS

This application claims priority of PCT International Application PCT/BR2004/000179, filed Sep. 20, 2004, which designated the United States, and is a continuation in part of provisional application Ser. No. 60/506,520, filed Sep. 26, 2003, provisional application Ser. No. 60/508,979, filed Oct. 6, 2003, provisional application Ser. No. 60/529,246, filed Dec. 12, 2003, and provisional application Ser. No. 60/560,466, filed Apr. 7, 2004, all of which are incorporated by reference in their entirety.

SEQUENCE LISTING

The instant application contains a Sequence Listing which has been submitted in ASCII format via EFS-Web and is hereby incorporated by reference in its entirety. Said ASCII copy, created on May 4, 2011, is named 05999401.txt and is 93,350 bytes in size.

FIELD OF THE INVENTION

The invention relates generally to the fields of molecular biology, biochemistry, plant pathology, and agriculture. More particularly, the invention relates to polynucleotides and proteins from a phytopathogenic virus, suitable for disease diagnosis and generation of transgenic plants with resistance to infectious viruses strains.

BACKGROUND OF THE INVENTION

Several Citrus diseases have been shown to be caused by infection with pathogenic viruses (Derrick, K. S. and Timmer, L. W., *Annu. Rev. Phytopathol.*, 38. 181-205 (2000)). One of the most important of these viruses is Citrus Tristeza Virus (CTV), a member of the Closterovirus group which induces serious disease syndromes in citrus. For example, CTV induces quick decline that causes the death of trees grafted on sour orange rootstock, and stem pitting of scion cultivars regardless of the rootstock used (Bar-Joseph et al., *Annu. Rev. Phytopathol.*, 27. 291-316 (1989)). These diseases cause significant losses in the citrus industry worldwide.

In Brazil, citrus tristeza, first detected in 1937, destroyed millions of trees of sweet orange grafted on sour orange rootstocks. The problem was solved by exchanging sour orange rootstock with Rangpur lime rootstock. Today, more than 85% of the 200 million citrus trees in Brazil are grafted on Rangpur lime rootstock (Gimenes-Fernandes, N. and Bassanezi, R. B., *Summa Phytopathologica*, 27. 93 (2001)).

In 1999, a new citrus disease, named Citrus Sudden Death (CSD) was discovered in Brazil (Gimenes-Fernandes, N. and Bassanezi, R. B. *Summa Phytopathologica*, 27. 93 (2001)). This disease affects sweet orange (*Citrus sinensis*) grafted on Rangpur lime rootstock (*Citrus limonia*), and causes the death of trees within a few months after the symptoms manifest (Gimenes-Fernandes, N. and Bassanezi, R. B. *Summa Phytopathologica*, 27. 93 (2001)). Although the disease was first observed in the sweet orange/Rangpur lime scion/rootstock combination, it has also been observed in orange trees cvs. Hamlin, Natal, Valencia, Pera, and Rubi, all grafted onto Rangpur lime (Bassanezi et al., *Phytopathol.*, 93. 4. 502-512 (2003)).

## 2

Plants with CSD symptoms present generalized leaf discoloration, partial defoliation, decreased number of young shoots and absence of internal shoots (Bassanezi et al., *Phytopathol.*, 93. 4. 502-512 (2003)). As the symptoms become more pronounced, the disease progresses rapidly and leads ultimately, to the death of the plant. The physiological status of the plant is important for the disease progression, since the severity of the symptoms increase at high water demand (Gimenes-Fernandes, N. and Bassanezi, R. B. *Summa Phytopathologica*, 27. 93 (2001)). The root system of the symptomatic plants is severely damaged and dies quickly as the disease progresses. CSD is also characterized by the development of a strong yellow stain in the phloem of the Rangpur lime rootstock (Gimenes-Fernandes, N. and Bassanezi, R. B. *Summa Phytopathologica*, 27. 93 (2001)). The time between the appearance of the first visible symptoms in the canopy and the death of the plant ranges from 1 to more than 12 months depending on season and citrus variety (Bassanezi et al., *Phytopathol.*, 93. 4. 502-512 (2003)).

The number of symptomatic trees in one affected area (north of São Paulo State and south of Triângulo Mineiro region, west of Minas Gerais State, Brazil), where the disease was originally found, increased from 500 in 1999 to more than 300,000 in February, 2002, and more than 1 million in June 2003 (Bassanezi, et al., *Phytopathol.*, 93. 4. 502-512 (2003)); (Román et al., *Plant Disease*, 88. 5. 453-467 (2004)). The pattern of CSD dissemination is similar to that of quick-decline, a disease caused by certain CTV isolates that elicit a graft union incompatibility when infected sweet orange scions are grafted onto sour orange rootstocks (Bassanezi et al., supra) however, CSD affects several sweet oranges grafted on Rangpur lime, a rootstock/scion combination that is not affected by the CTV strains that causes quick-decline (Bassanezi et al., supra).

Based on the spatial and temporal patterns of CSD dissemination, it has been hypothesized that CSD may be caused by an insect-vector pathogen, potentially a new, undescribed strain of CTV (Bassanezi et al., supra.) Alternatively, a new virus could be the causative agent of the CSD disease.

To test if CSD is caused by a variant strain of CTV or is caused by a new virus, a genomic approach using shotgun sequencing of genomic viral RNA that had been randomly reverse transcribed and cloned in a plasmid vector, was used to study the disease described herein. Bioinformatic tools were developed for the identification and assembly of viral sequences. Using this approach, it was possible to obtain a saturated database of viral sequences from individual trees.

Genomic viral RNA isolated from citrus trees symptomatic or asymptomatic for CSD were reverse transcribed and the first strand cDNA was used to construct random-primed cDNA libraries. Around 2,000 cDNA clones from each tree were sequenced and the sequences were analyzed using BLASTX, BLASTN, and TBLASTX searches against public databases. A viral genome assembled consensus sequence of 6820 nucleotides (SEQ ID NO: 1) encoding a viral polyprotein (SEQ ID NO: 2) sufficient to assemble a viral particle, was identified.

The 6820 nucleotides viral genome sequence, when translated in all possible frames, give rise to, at least, the following polypeptides: a Major Capsid Protein (Coat Protein 1) encoded by the Nucleotide Sequence Domain (NSD1) starting at nucleotide position 6028 and ending at nucleotide position 6675 of SEQ ID NO: 1, whose translation product give rise to the Amino Acid Sequence Domain (AASD) of the polypeptide of SEQ ID NO: 2, starting, at amino acid position 1974 and ending at amino acid position 2188; a Minor Capsid Protein (Coat Protein 2) encoded by NSD2 of SEQ ID NO: 1

starting at nucleotide position 6082 and ending at nucleotide position 6675, whose translation product give rise to the AASD of the polypeptide of SEQ ID NO: 2, starting at amino acid position 1992 and ending at amino acid position 2188; a Putative Movement Protein encoded by NSD3 of SEQ ID NO: 1 starting at nucleotide position 6260 and ending at nucleotide position 6724, whose translation product give rise to the AASD of the polypeptide of SEQ ID NO: 3, starting at amino acid position 1 and ending at amino acid position 154; a Methyltransferase Domain encoded by NSD4 of SEQ ID NO: 1 starting at nucleotide position 487 and ending at nucleotide position 1119, whose translation product give rise to the AASD of the polypeptide of SEQ ID NO: 2, starting at amino acid position 127 and ending at amino acid position 337; a Protease Domain encoded by NSD5 of SEQ ID NO: 1 starting at nucleotide position 2797 and ending at nucleotide position 3114, whose translation product give rise to the AASD of the polypeptide of SEQ ID NO: 2, starting at amino acid position 897 and ending at amino acid position 1002; a Helicase Domain encoded by NSD6 of SEQ ID NO: 1 starting at nucleotide position 3358 and ending at nucleotide position 4053, whose translation product give rise to the AASD of the polypeptide of SEQ ID NO: 2, starting at amino acid position 1084 and ending at amino acid position 1315; a RNA-dependent RNA polymerase encoded by NSD7 of SEQ ID NO: 1 starting at nucleotide position 4528 and ending at nucleotide position 5778, whose translation product give rise to the AASD of the polypeptide of SEQ ID NO: 2, starting at amino acid position 1474 and ending at amino acid position 1890.

The viral genome sequence showed strong similarity to several viruses from the Tymoviridae family of plant viruses, especially the oat blue dwarf virus (FIGS. 1 and 2). Analysis of CSD-symptomatic or asymptomatic trees for the presence of these viral sequences revealed that only trees presenting the CSD symptoms contain the viral sequences. It was therefore assumed that these sequences belong to an undescribed virus of the Tymoviridae family which is the causative agent of the CSD disease.

#### SUMMARY OF THE INVENTION

The present invention relates to nucleic acid molecules of and from the genome of a virus that is the causative agent of CSD disease. Sequence comparison with a number of viral genome sequences revealed that this new virus belongs to the Tymoviridae family. This new virus is herein named "Citrus Sudden Death Virus," or "CSDV". It is hereafter an object of the invention to provide nucleic acid molecules which encode infectious CSDV. Such nucleic acid molecule is referred to throughout the application as "CSDV nucleic acid molecules".

For the purposes of this application, nucleic acid molecules refers to RNA, DNA, cDNA or any variant thereof with functions equivalent to RNA, gDNA, and cDNA, such as the synthesis of CSDV polypeptide domains. Also, the polypeptide domains encoded by CSDV nucleic acid molecules are referred to throughout the application as "CSDV polypeptides" or "CSDV proteins".

The invention relates to the use of the CSDV nucleic acid molecules to produce polypeptides. "Nucleic acid molecules of the invention" refers to, e.g., CSDV nucleic acid molecules, mutations of CSDV nucleic acid molecules, chimeric nucleic acid molecules and so forth.

In one embodiment, polypeptides are produced by cells transfected with nucleic acid molecules of the invention. In another embodiment, the polypeptide or polypeptides are produced recombinantly from a fragment or portion of the

nucleic acid molecules of the invention. In yet another embodiment, the polypeptides are chemically synthesized.

Since, in nature, the CSDV proteins are ultimately synthesized from the information contained in the genome sequence of CSDV, the invention also relates to the use of the CSDV particles isolated and purified from infected plants tissues and/or organs. In one embodiment the particles can be used to produce antibodies against the CSDV proteins. In another embodiment the purified CSDV particles can be used to isolate and purify CSDV protein domains that can further be used to produce antibodies against CSDV.

The polypeptides of the invention can serve, e.g., as immunogens in the development of diagnostic assays for detecting the presence of CSDV in biological samples, as they provoke antibody production, and the antibodies can then be used in assays.

The invention also relates to the use of the CSDV nucleic acid molecules for diagnosis purposes, in which oligonucleotide primers containing from 5 to 100 nucleotides presenting from 90 to 100% identity with the CSDV nucleotide sequences can be used in, e.g., RT-PCR reactions, so that parts of the CSDV nucleic acid molecules can be amplified and detected in ordinary agarose gels serving as a diagnostic for the presence of the virus.

The invention also relates to methods of transforming plants, such as monocots or dicots, with constructs containing the CSDV nucleic acid molecules, to produce plants that are resistant to CSDV. Such methods include the introduction of constructs containing at least one CSDV nucleic acid molecule into plant parts, such as scions, rootstock cultivars, and so forth, as well as into citrus germplasm and breeding lines. Transformed CSDV-resistant germplasm and breeding lines can be used in conventional breeding programs, to create new cultivars that carry and express the resistance genes.

Accordingly, the invention features (i) isolated and/or purified CSDV nucleic acid molecules that have at least 65% sequence identity with the nucleotide sequence of SEQ ID NO.: 1; (ii) sequences complementary thereto or nucleotides whose complement hybridizes under high stringency conditions to the nucleotide sequence of SEQ ID NO.: 1; as well as with the coding regions of the domains encoded therein (iii) polypeptides or portions of polypeptides that have at least 70%, more preferably 80% sequence identity with the amino acid sequence of SEQ ID NO.: 2, or with any of the domains within SEQ ID NO:2 as defined herein.

"Stringent conditions" as used herein, refers to parameters with which the art is familiar, i.e., hybridization in 3.5×SSC, 1×Denhardt's solution, 25 mM sodium phosphate buffer (pH 7.0), 0.5% SDS, and 2 mM EDTA for 18 hours at 65° C., followed by 4 washes of the filter at 65° C. for 20 minutes, in 2×SSC, 0.1% SDS, and a final wash for up to 20 minutes in 0.5×SSC, 0.1% SDS, or 0.3×SSC and 0.1% SDS for greater stringency, and 0.1×SSC, 0.1% SDS for even greater stringency. Other conditions may be substituted, as long as the degree of stringency is equal to that provided herein, using a 0.5×SSC final wash.

The invention also features expression vectors or constructs in which the CSDV nucleic acid molecules are operably linked to one or more expression control sequences or promoters.

The invention further features a transgenic plant, such as one of the genera *Citrus* and *Poncirus*, into which a CSDV nucleic acid molecule has been introduced. Exemplary are citrus scions and rootstock cultivars (e.g., from sour orange [Citrus aurantium], Rangpur lime [Citrus limonia], rough lemon [Citrus limonia and Citrus jambhiri], mandarin "Cleopatra" [Citrus reshni], Sunki [Citrus sunki], Volkamerian

lemon [*Citrus volkameriana*], “Caipira” orange [*Citrus sinensis*] and intrageneric hybrids (e.g., tangelos [*Citrus paradisi*]*xCitrus reticulata*], tangors [*Citrus reticulata*]*xCitrus sinensis*], citrumelo [*Poncirus trifoliata*]*xCitrus maxima*], and citrange [*Citrus sinensis*]*xPoncirus trifoliata*]). The plant can be a breeding line. It can also be one from *Fortunella* and *Citrofortunella* species, including calamondin and kumquat. The nucleic acid molecule in the plant preferably includes a selectable marker such as an herbicide resistance gene.

The invention relates to nucleic acid molecules, and the polypeptides they encode, which were identified using shotgun sequencing of viruses genomes from citrus plants with or without symptoms of CSD. These sequences come from a newly identified virus of the Tymoviridae family which, when it infects a plant, such as a citrus plant, causes CSD disease, leading to the death of the plant. Methods of genetic transformation to produce plants that are resistant to CSDV strains are within the invention. Such methods include the introduction of constructs containing the CSDV nucleic acid molecules into scions and/or rootstocks, so that commercial varieties or any other germplasm useful for breeding programs can be produced and used to create new cultivars resistant to CSDV.

Accordingly, the invention involves purified CSDV nucleic acid molecules, which may be isolated from citrus plants manifesting symptoms of CSD, that when used in appropriate constructs have the ability to confer resistance to pathologies caused by CSDV infection in plants infected with CSDV. The nucleic acid molecule can be a purified portion or a gene present in the complete genome of CSDV, e.g., a nucleic acid molecule whose nucleotide sequence encodes a polypeptide or a portion of a polypeptide that has at least 80% sequence identity to the amino acid sequence of SEQ ID NO.: 2. The nucleotide sequences can be that of SEQ ID NO.: 1 or one that defines polynucleotides whose complement hybridizes under high stringency conditions to the nucleotide sequences of SEQ ID NO.: 1.

The invention also features expression vectors, isolated recombinant cells, and plants and plant parts containing the nucleic acid molecule of the invention, e.g., one of the nucleic acid molecules described above. The nucleic acid molecule in the vector, cell, seed or plant can be operably linked to one or more expression control sequences or promoters.

In addition, the invention features the purified proteins encoded by the nucleic acid sequence domains present in the genome sequence of CSDV. The proteins include those with amino acid sequences that (a) share at least 80% sequence identity with SEQ ID NO.: 2 or (b) includes the amino acid sequences of SEQ ID NO.: 2. Proteins of the invention can be expressed in bacteria either as “neat” proteins, or as heterologous or fusion polypeptides. The invention also features antibodies raised against the proteins of the invention, e.g., those described above. The antibodies can further include a detectable label.

The invention also features a plant transfected with one of the nucleic acid molecules of the invention, e.g., any purified CSDV sequence. The plant may be of the genera *Citrus* and *Poncirus* (e.g., sweet orange [*Citrus sinensis*], mandarin [*Citrus reticulata*], sour orange [*Citrus aurantium*], Rangpur lime [*Citrus limonia*], rough lemon [*Citrus limonia* and *Citrus jambhin*], mandarin “Cleopatra” [*Citrus reshni*], Sunki [*Citrus sunki*], Volkamerian lemon [*Citrus volkameriana*], “Caipira” orange [*Citrus sinensis*]) and intrageneric hybrids (e.g., tangelos [*Citrus paradisi*]*xCitrus reticulata*], tangors [*Citrus reticulata*]*xCitrus sinensis*], citrumelo [*Poncirus trifoliata*]*xCitrus maxima*], and citrange [*Citrus sinensis*]*xPon-*

*cirus trifoliata*]). The plant can be a breeding line. It can also be one from *Fortunella* and *Citrofortunella* species, including calamondin and kumquat.

The invention features a method of producing a disease resistant plant by introducing constructs containing purified nucleic molecules under the control of a suitable plant promoter, into the plant, transforming the plant with *Agrobacterium* strains or microprojectile bombardment.

All technical terms used herein are terms commonly used

- 10 in biochemistry, molecular biology, phytopathology and agriculture, and will be understood by one of ordinary skill in the art to which this invention belongs. Those technical terms can be found in, e.g., Molecular Cloning: A Laboratory Manual, 3rd ed., vol. 1-3, ed. Sambrook and Russel, Cold Spring Harbor Laboratory Press, Cold Spring Harbor, N.Y., 2001; Current Protocols in Molecular Biology, ed. Ausubel et al., Greene Publishing Associates and Wiley-Interscience, New York, 1988 (with periodic updates); Short Protocols in Molecular Biology: A Compendium of Methods from Current Protocols in Molecular Biology, 5<sup>th</sup> ed., vol. 1-2, ed. Ausubel et al., John Wiley & Sons, Inc., 2002; Genome Analysis: A Laboratory Manual, vol. 1-2, ed. Green et al., Cold Spring Harbor Laboratory Press, Cold Spring Harbor, N.Y., 1997. Methods involving plant biology techniques are
- 15 described herein and are described in detail in methodology treatises such as Methods in Plant Molecular Biology: A Laboratory Course Manual, ed. Maliga et al., Cold Spring Harbor Laboratory Press, Cold Spring Harbor, N.Y., 1995. Various techniques using PCR are described, e.g., in Innis et al., PCR Protocols: A Guide to Methods and Applications, Academic Press, San Diego, 1990 and in Dieffenbach and Dveksler, PCR Primer: A Laboratory Manual, 2<sup>nd</sup> ed., Cold Spring Harbor Laboratory Press, Cold Spring Harbor, N.Y., 2003. PCR-primer pairs can be derived from known
- 20 sequences by known techniques such as using computer programs intended for that purpose (e.g., Primer, Version 0.5, 1991, Whitehead Institute for Biomedical Research, Cambridge, Mass.). Methods for chemical synthesis of nucleic acids are discussed, for example, in Beauchage and Caruthers (1981) *Tetra. Letts.*, 22:1859-1862 and Matteucci and Caruthers (1981) *J. Am. Chem. Soc.*, 103:3185.

#### BRIEF DESCRIPTION OF THE DRAWINGS

45 The invention can be more readily understood by reference to the accompanying drawings, wherein:

FIG. 1 shows the alignment of the nucleotide sequence of the CSDV genome (SEQ ID NO: 1) with that of the Tymoviridae Oat Blue Dwarf Virus/OBDV-GI 9629255 (SEQ ID NO: 13).

50 FIG. 2 shows the alignments of the amino acid sequence of the polyprotein encoded by the CSDV genome (SEQ ID NO: 2) with those encoded by other Tymoviridae genomes [Oat Blue Dwarf Virus/OBDV—GI 9629255 (SEQ ID NO: 14), 55 Grapevine Fleck Virus/GFkV—GI 18138525 (SEQ ID NO: 19), Grapevine Asteroid Mosaic-Associated Virus/GAMaV—GI 29335718 (SEQ ID NO: 15)]. FIG. 2 (b) discloses SEQ ID NOs: 15-16, respectively, in order of appearance. FIG. 2 (c) discloses SEQ ID NOs: 17-18, respectively, 60 in order of appearance. FIG. 2 (d) discloses SEQ ID NOs: 19 and 3, respectively, in order of appearance.

FIG. 3 shows the PCR amplification of a DNA fragment amplified with primers based on the CSDV nucleotide sequence of SEQ ID NO.: 1. CDNA samples were prepared 65 from symptomatic and asymptomatic citrus trees. Only trees presenting the CSD symptoms contain an amplifiable CSDV sequence of SEQ ID NO.: 1.

FIG. 4 shows the results of a Northern Blot experiment in which RNA samples from symptomatic and asymptomatic plants were subjected to an RNA gel blot analysis to verify the presence of RNA species able to hybridize with a DNA probe derived from the CSDV sequences. Only lanes containing RNA samples from symptomatic trees possess a band sizing 6.5 kbp representing the complete CSDV viral RNA genome.

FIG. 5 depicts the results of a RT-PCR experiment performed on total RNA taken from insect bodies, to determine the vectors that transmit CSD.

FIG. 6 shows the results of a western blot experiment in which total protein extract from symptomatic and asymptomatic plants where subject to a protein gel blot analysis developed with antibody against the CSDV Coat Proteins 1 and 2, respectively.

FIG. 7 shows an electron micrography of virus particles purified from citrus plants presenting CSD symptoms.

FIG. 8 shows a Northern blot of RNA extract from purified virus particle.

FIG. 9 shows a drawing of the vector pTYMO-AS that contain part of the RNA-dependent RNA polymerase sequence domain (SD7, as defined in SEQ NO.: 1) operably linked to the CaMV 35S promoter. This vector was used to transform citrus plants by *Agrobacterium* mediated transformation.

#### DETAILED DESCRIPTION OF THE INVENTION

Nucleic acid molecules from the genome of an undescribed virus that causes Citrus Sudden Death (CSD) have been cloned and sequenced. Such nucleic acid molecules are referred to throughout the application as "CSDV nucleic acid molecules". Polypeptides predicted from CSDV nucleic acid molecules have been analyzed using software programs including BLAST, and have been shown to encode, inter alia, an RNA polymerase, a methyltransferase, a protease and a helicase that are involved in the replication of the CSDV, a movement protein involved in the translocation of the virus throughout the plant, and the capsid proteins responsible for the encapsulation of the virus genome.

The molecular cloning of CSDV nucleic acid molecules provides the means to develop diagnostic methods to detect the presence of CSDV in biological samples, including tissues, cells and organs of plants, such as plants of the genus *Citrus*. The molecular cloning of CSDV nucleic acid molecules also provides the means to create CSD-resistant plants of the genus *Citrus* through genetic transformation. Genetic transformation of plants of the genus *Citrus*, can be obtained using *Agrobacterium* mediated transformation methods. Such methods include cloning constructs containing CSDV nucleic acid molecules operably ligated to promoter and enhancer regions, initiation and termination sequences. These constructs can also contain genes for selectable markers, such as herbicide resistance. These constructs may be cloned in the Ti plasmid of *Agrobacterium*. Plasmid vector-containing constructs are used to transform commonly used *Agrobacterium* strains, which are subsequently used to transform plants, such as those of the genus *Citrus*. Plasmid vector-containing constructs may be also introduced into plants by microprojectile bombardment. The constructs containing the CSDV nucleic acid molecules are useful for creating CSD resistant plants such as all common types of citrus fruits, including but not limited to sweet oranges, grapefruit, mandarins, tangerines, pummelos, lemons, limes, citrons, bergamots, limequats, meyer lemons, silver limes, key limes, kaffir limes, lavender gems, blood oranges, satsumas, oroblanços, melogolds, bergamots, intrageneric hybrids such as tangelos

and tangors, and citrus-type fruit such as calamondins and kumquats (*Fortunella* spp.). For example, CSDV nucleic acid molecules can be introduced into commercially utilized rootstock cultivars, including but not been limited to, Rangpur lime, sour orange, rough lemon, various mandarins, and citrus intrageneric and intergeneric hybrids. CSD resistant citrus plants, composed of genetic modified scions and rootstocks, can then be used by citrus growers to counter CSD disease, and to avoid decreased productivity and/or tree death and 10 replanting costs.

The invention provides purified nucleic acid molecules (polynucleotides) that encode polypeptides having an amino acid sequence such as that of SEQ ID NO.: 2.

The CSDV nucleic acid molecules of the present invention 15 can be obtained from CSDV infected plants. The molecules of the present invention may be in the form of RNA or in the form of cDNA. The cDNA may be double- or single-stranded, and, if single-stranded may be the coding (sense) strand or noncoding (anti-sense) strand. The sequence may be identical 20 to a nucleotide sequence of SEQ ID NO.: 1. It may also be a different coding sequence which, as a result of the redundancy or degeneracy of the genetic code, encodes the same polypeptide as polynucleotides of SEQ ID NO.: 1. Other nucleic acid molecules within the invention are variants of 25 CSDV nucleic acid molecules such as those that encode fragments, analogs and derivatives of native CSDV nucleic acid molecules. Such variants may be, e.g., naturally occurring polymorphic variants of native CSDV nucleic acid molecules, or a non-naturally occurring variant of native CSDV 30 nucleic acid molecules. For example, the nucleotide sequence of such variants can feature a deletion, addition, or substitution of one or more nucleotides of native CSDV nucleic acid molecules. Naturally occurring variants of native CSDV nucleic acid molecules within the invention are nucleic acids 35 isolated from CSDV infected plants that have at least 65% sequence identity with native CSDV nucleic acid molecules, and encode polypeptides having at least one functional activity in common with native CSDV nucleic acid molecules encoded proteins.

40 Shorter oligonucleotides (e.g., those from 6-200, preferably 12-200, more preferably 20, 30, or 50 to 200 (100, 125, 150 or 200) base pairs in length), e.g., that match perfectly to the CSDV nucleic acid molecules or hybridize with CSDV nucleic acid molecules at stringent conditions as defined 45 herein can be used as probes, primers, or antisense molecules.

Longer polynucleotides (e.g., those of 300 to 800 base pairs) that encode or hybridize with CSDV nucleic acid molecules, can be used in place of a native CSDV nucleic acid molecule in applications where it is desired to modulate the 50 functional activity of a native CSDV nucleic acid molecule.

Nucleic acids molecules that hybridize under stringent conditions as defined herein to CSDV nucleic acid molecules of SEQ ID NO.: 1 or the complement of the SEQ ID NO.: 1 are also within the invention. For example, such nucleic acids 55 can be those that hybridize with the CSDV nucleic acid molecules of SEQ ID NO.: 1 or the complement of the SEQ ID NO.: 1, under low stringency conditions, moderate stringency conditions, or high stringency conditions, and are within the scope of the invention. Preferred nucleic acids molecules are 60 those having a nucleotide sequence that is the complement of all or a portion of the CSDV nucleic acid molecules of SEQ ID NO.: 1. Other variants of nucleic acid molecules within the invention are polynucleotides that share at least 65% sequence identity to the CSDV nucleic acid molecules of SEQ ID NO.: 1 or the complement of SEQ ID NO.: 1.

Other CSDV nucleic acid molecules encoding polypeptides are also within the invention. Such polypeptides can be

made by preparing a construct (e.g., an expression vector) that expresses CSDV nucleic acid molecules encoding polypeptides, when introduced into a suitable host. Variant CSDV nucleic acid molecule-encoding polypeptides can be produced by those skilled in molecular biology procedures using standard nucleic acid mutagenesis techniques or chemical synthesis, or the polypeptides can be isolated and purified from CSDV particles isolated and purified from infected plants, as the CSDV particles encoded by nucleic acid molecules of SEQ ID NO.: 1 and polypeptide domains of SEQ ID NO.: 2. Antibodies can be produced against the isolated and purified CSDV particles and can be used for serological diagnosis of the virus.

Another aspect of the invention relates to the use of purified antisense nucleic acids to inhibit expression of CSDV nucleic acid molecules. Antisense nucleic acid molecules within the invention are those that specifically hybridize under cellular conditions to cellular mRNA and/or genomic RNA of CSDV in a manner that inhibits expression of the nucleic acid domains encoded by the CSDV nucleic acid molecules.

The antisense nucleic acid molecules can be delivered into cells that express CSDV genes. For instance, constructs expressing antisense molecules under the control of a strong promoter can be introduced into citrus plants by genetic transformation using *Agrobacterium* or microprojectile bombardment (Ghorbel et al., *Tree Physiology*, 20, 1183-1189 (2000); Bespalhok et al., *Crop Breed. Appl. Biotech.*, 1, 27-34 (2001); Bespalhok et al., *Braz. Arch. Biol. Technol.*, 46, 1, 1-6 (2003); Molinari et al., *Scientia Horticulturae*, 99, 34, 379-385 (2004); Jia-Long et al., *Plant Science*, 113, 2, 175-183 (1996)).

The expression of CSDV nucleic acid molecules can be modulated by RNA interference (RNAi) (Lee et al. *Nature Biotech.*, 19, 500-505 (2002); Voinnet, O. *Trends Genet.*, 17, 449-459 (2001)) by which a construct driving the synthesis of sequence-specific double-stranded RNA (dsRNA) is introduced into an organism or cell in order to silence the targeted gene (Hannon, *Nature*, 418, 244-251 (2002)). Selected sequences corresponding to CSDV nucleic acid molecules can be used to create, after expression, a sequence-specific dsRNA that can interfere with accumulation of endogenous RNA encoded by the CSDV nucleic acid molecules.

The CSDV nucleic acid molecule can be altered by using molecular biology techniques to produce a mutant recombinant virus that work as a vaccine. This is well known in the art as cross-protection system, in which a mild, non-infective virus is introduced in a plant and after replication induces in the host plant a defense response against the severe, infective strain (Costa, A. S. and Muller, G. W. 1980, *Plant Dis.*, 64:538-541). Plants inoculated with recombinant CSDV mutant become resistant to the wild-type CSDV infective strains.

The present invention is further illustrated by the following specific examples. The examples are provided for illustration only and are not to be construed as limiting the scope or content of the invention in any way.

## EXAMPLES

### Example 1

This example describes the identification and cloning of nucleic acid molecules corresponding to the complete genome sequence of CSDV. cDNA libraries were constructed from double strand RNA isolated from citrus plants presenting symptoms of CSD by shotgun cloning of cDNAs generated by RT-PCR. Clones were randomly sequenced using an

ABI 3700 sequencer. Sequences were trimmed for vector bases and low quality bases and BLASTX-analyzed against the non-redundant (NR) GenBank database. Two cDNA clones were identified as having nucleotide sequences similar to viral nucleotide sequences of the Tymoviridae family. By primer walking PCR using a cDNA library or total RNA from CSD plants as templates, cDNA clones were identified that, after sequencing on both ends, gave rise to a consensus CSDV nucleic acid sequence of 6820 nucleotides. This consensus sequence contains nucleotide sequence domains that encode the complete set of the viral proteins comprised of: a Major Capsid Protein (Coat Protein 1), a Minor Capsid Protein (Coat Protein 2), a Putative Movement Protein, a Methyltransferase Domain, a Protease Domain, a Helicase Domain and a RNA-dependent RNA polymerase. The gene and protein domains organization of CSDV is similar to that found in several virus strains of the Tymoviridae family, especially to the Oat blue dwarf virus (FIGS. 1 and 2).

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### Example 2

This example describes RT-PCR analysis of citrus plants to determine if the CSDV nucleic acid molecules of the invention could be used to design oligonucleotide primers that amplify CSDV sequences and could be use in diagnostic assays. Oligonucleotide primers designed on the basis of CSDV nucleic acid molecules of SEQ ID NO.: 1 were used to amplify the CSDV nucleic acid sequences using RT-PCR from RNA isolated from CSD symptomatic or asymptomatic citrus plants. Bark from young citrus branches was peeled and ground to a powder, in liquid nitrogen. Total RNA was purified from 100 mg of bark tissue by using Trizol reagent according to the manufacturer's instructions. The resulting total RNA was suspended in 50 ul of DEPC (diethylpyrocarbonate)-treated sterile water and used for cDNA synthesis. First strand cDNA was synthesized using 8 ul of total RNA (approx. 8 ug) as template. Two microliters of random primers (1 ug) were added to the total RNA that had been denatured at 97° C. for 5 min. The solution was then incubated on ice while adding 1 ul of 10 mM dNTP mix and 3 ul of First Strand buffer (250 mM Tris-HCl, pH 8.3, 375 mM KCl, 15 mM MgCl<sub>2</sub>) to the tube. The mixture was incubated at room temperature for 2 min, 1 ul (200 U) of the enzyme reverse transcriptase SuperScript II was added and the solution was further incubated at room temperature for 10 min. followed by 60 min. at 42° C. For PCR, 1 ul of the synthesized cDNA was added to a 20 ul reaction containing 0.5 mM of each of primers "TYMOF2" 5'-GTCAGCTGTCCAACCAGTCC-3' (SEQ ID NO: 4) and "TYMORR": 5'-GTGAAGATCAAT-GAGAGCCTG-3'(SEQ ID NO: 5), 0.125 mM each dNTP, 2.5 mM MgCl<sub>2</sub>, 1x reaction buffer (20 Mm Tris-HCl, pH 8.4, 50 mM KCl), and 1 U of Taq polymerase. The reaction was heated for 2 min. at 94° C. and subjected to 40 amplification cycles (30 s at 94° C., 30 s at 55° C., 1 min at 72° C.). Ten microliters of the RT-PCR reaction were combined with 10 ul of 2x digestion buffer (100 mM potassium acetate, 40 mM Tris-acetate, 20 mM magnesium acetate, 2 mM DTT and 4 ug BSA, pH 7.9) and 0.5 ul of the restriction enzyme Apal (5 U). Digestion was carried out for 2 hours at 25° C. DNA fragments were separated in a 1.5% agarose gel, stained with 100 ng/ml ethidium bromide and compared under UV light.

The results are depicted in FIG. 3. Lanes a-f and m-t are from asymptomatic plants, while g to l, and u to z are from symptomatic plants. There is no banding at all in lanes a-f and m-t, while clear banding is evident in the samples from symptomatic plants in lanes g to l and u to z, which generate fragments of 500 and 250 base pairs long when digested with

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Apal enzyme. Data was further expanded to 512 plants (351 symptomatic and 161 asymptomatic) and 99.7% of yellow bark plants shows the CSDV nucleic acid fragment amplification.

## Example 3

In these experiments, Northern blotting was carried out on samples of RNA taken from asymptomatic and symptomatic plants. A DNA probe taken from CSDV nucleic acid molecules was used. FIG. 4 shows that a band of 6.5 kbp was identified in symptomatic plants, while no banding was identified in any of the asymptomatic material.

Total RNA from bark tissue of symptomatic and asymptomatic plants was extracted using the Trizol Reagent (Invitrogen) according to the manufacturer's protocol. Fifteen micrograms of total RNA from each sample were separated by denaturing electrophoresis on a 1% agarose gel containing 1×MOPS and 0.6M formaldehyde, according to Sambrook and Russell (Molecular Cloning: A Laboratory Manual, 3rd ed., vol. 1-3, ed. Cold Spring Harbor Laboratory Press, Cold Spring Harbor, N.Y., 2001). The gel was subsequently transferred to a Hybond-N+ nylon membrane (Amersham Pharmacia Biotech) by capillary transfer in 10×SSC (1×SSC is 0.15M NaCl; 0.015M sodium citrate) for 16 h. The membrane was baked at 80°C. for 2 h, prehybridized in ExpressHyb hybridization buffer (Clontech) for 2 h at 65°C., followed by hybridization in a fresh aliquot of ExpressHyb solution containing 20 ng/ml of probe (2×10<sup>7</sup> cpm/ml) for 16 h at 65°C. The probe consisted of a 772-bp CSDV nucleic acid fragment amplified by PCR from a plasmid containing part of SEQ ID NO.: 1 using SEQ ID NOS: 4 and 5. The probe was radioactively labeled by random-priming with [ $\alpha$ -32P]dCTP (6000 Ci/mmol) using the Random Primers DNA Labeling System (Invitrogen). After hybridization, the membrane was washed at room temperature in 2×SSC, 0.05% SDS for 4×10 min, and then twice at 55°C. for 20 min each in 0.1×SSC; 0.1% SDS. The blot was exposed for three days and analyzed by phosphoimaging on a FLA-3000 Fluorescent Image Analyzer (Fujifilm).

## Example 4

This example describes RT-PCR analysis of citrus insect vectors to determine if the CSDV nucleic acid molecules of the invention could be found in insect vectors and therefore determine the kind and species of insects that could act as a vector for transmission and dissemination of CSD in citrus plantations.

A number of insect species including aphids such as *Toxoptera citricida*, *Aphis spiraecola* and *Aphis gossypii*, and leafhoppers/planthoppers representing, but not limited to, *Deois flavopicta*, *Xerophloea viridis*, *Ferrariana trivittata*, *Hortensia similis*, *Erytrogonia sexguttata*, *Macugonalia leucomelas*, *Dechacona missionum*, and *Copididonus hialipennis* were sampled at citrus plantation areas affected by CSD and at areas without symptoms of CSD. In the areas affected by CSD the insects were collected from individual trees presenting symptoms of CSD. Leafhoppers/planthoppers were separated into 6 groups according to morphological characteristics, while aphids were separated according the respective species. Around fifty individuals of each leafhoppers/planthoppers group or aphids species were used to extract total RNA.

Total RNA from whole bodies of insects was extracted using the Trizol Reagent (Invitrogen) according to the manufacturer's protocol. Eight micrograms of total RNA from

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each sample were used for RT-PCR analysis using CSDV specific primers designed on the basis of the CSDV nucleic acid molecules of SEQ ID NO: 1.

RT-PCR was carried out on samples of total RNA using SEQ ID NOS: 4 and 5. FIG. 5 shows that a band of 772 Kbp corresponding to part of the CSDV nucleic acid domain encoding the RNA-dependent RNA polymerase was amplified from samples taken from the aphids *Toxoptera citricida*, *Aphis spiraecola* and *Aphis gossypii* collected from symptomatic trees in the affected areas, while no band was amplified in any of the RNA samples isolated from leafhoppers or planthoppers of CSD affected areas, or from aphids collected in trees from the non-affected areas.

Table 1 presents a summary of the results of the assays for the presence of CSDV in samples of insects collected in CSD affected and non-affected areas. The virus was found only in the aphid samples from citrus trees of CSD affected areas. This suggests that these aphids could act as vectors for CSDV transmission and dissemination.

## Example 5

This example illustrate the use of polyclonal antibodies raised against the CSDV Coat Proteins 1 and 2 to determine the presence of CSDV in plant extracts. The polyclonal antibodies was produced using two different sources of immunogens: 1) Peptides were designed based on the deduced amino acid sequences of Coat Proteins 1 and 2 of CSDV, taking into consideration b-turn structure prediction (Chou, P Y and Fasman, Biophys J, 1979 June; 26(3):367-73) and hydrophobicity level (Kyte, J. and Doolittle, R F, J Mol Biol., 1982 May 5; 157(1):105-32). Peptides corresponding to amino acids 7 to 20 (AGPAPSRRDRVDRQ (SEQ ID NO: 8)), 11 to 24 (PSRDDRVRDRQPRRLP (SEQ ID NO: 9)), 51 to 64 (DG-SEAKNLSDDLSG (SEQ ID NO: 10)), 111 to 124 (PASASETSYYGGRL (SEQ ID NO: 11)), and 161 to 175 (RFSYSVSYNSNGGTKG (SEQ ID NO: 12)) of the predicted Coat Protein 2 (amino acid positions 1974-2188 of SEQ ID NO: 2) were synthesized, KLH-conjugated (Genscript Corp, USA) and used to immunize rabbits. Polyclonal antibodies were delivered as antisera and used without further treatment for Western blot analyses. 2) Polyclonal antibodies were also obtained against the recombinant Coat Proteins 1 and 2. The CSDV nucleic acid sequences between nucleotide positions 6,028 to 6,672 (Coat Protein 1) and 6,082 to 6,672 (Coat Protein 2) were cloned into the pET-28a(+) expression vector (ClonTech) and expressed in *E. coli* BL21(DE3) strain. Coat Protein 1 was expressed as a fusion with a His-tag and thrombin site in the N-terminal (His-p22.5) and Coat Protein 2 was expressed fused to a His-tag in the C-terminal (p21-His) and also in a non-fused form (Nfp21). Large scale expression was conducted under IPTG induction, bacterial cells were disrupted by French press, and the soluble fraction of the fused recombinant proteins were purified using the His-Trap purification Kit (Amersham). Purified p21-His and Hisp22.5 were used to raise polyclonal antibodies in rabbits.

Recombinant CSDV Coat Proteins and crude protein extracts from citrus trees were resolved in a 12% (w/v) acrylamide gel and proteins were transferred to nitrocellulose membrane by semi-dry electroblotting (BioRad apparatus). The membrane was blocked with a Blotto solution (PBS, pH 7.2 containing 0.1% Tween-20 and 5% (w/v) non-fat-milk powder), incubated with the polyclonal antibodies (1:1000 in PBS) against the peptides, washed with PBS-Tween 20 and incubated with anti-rabbit IgG alkaline phosphatase conjugate (BioRad, 1:2000 in PBS). Color development was carried out in freshly prepared substrate solution (1.5 mg NBT, 3

mg BCIP in 20 ml carbonate buffer, pH 9.2). Antisera obtained for the peptides were able to recognize both recombinant Coat Proteins as well as Coat Proteins present in the CSDV-infected plant extracts (FIG. 6). Two protein bands of expected size were evident only in infected plant extracts and may correspond to Coat Proteins 1 and 2, respectively. Similar results were obtained using antibody raised against the recombinant CSDV Coat Proteins.

#### Example 6

In this example, CSDV particles were purified from CSD-affected citrus plants according to the methodology described by Bar-Joseph et al. (Bar-Joseph, M., D. J. Gumpf, J. A. Dodds A. Rosner, and I. Ginsberg. 1985, *Phytopathology*, 75:195-198.). The purity of virus preparation was determined by electron microscopy using a negative staining methodology with 2% uranyl acetate (Gaméz, R., T. Fukuoka, and Y. Kozuka. 1977, *Rev. Biol. Trop.*, 25:151-157). The electron micrograph shows purified virions as expected isometric, non-enveloped, ~30 nm in diameter particles, with a rounded contour, and prominent surface structure. As typical Tymovirus, CSDV present under uranyl acetate staining two types of particles (Boulila, M., D. Boscia, B. Di Terlizzi, M. A. Castellano, A. Minafra, V. Savino, and G. P. Martelli. 1990, *J. Phytopathol.*, 129:151-158), one with a negative stain (T form, represents non-infectious empty shells) and another with positive stain (B form, represents intact particles).

In order to confirm that the purified particles correspond to CSDV, total nucleic acids were purified from a preparation of virus particles by phenol extraction, resolved in a 1% agarose gel, transferred to Hybond-N+ nylon membranes (Amersham) and hybridized with a P<sup>32</sup>-labelled DNA probe from part of the CSDV nucleic acid molecules of SEQ ID NO.: 1. The results of FIG. 8, confirm the presence of the CSDV genomic RNA in the purified sample.

#### Example 7

This is an example of a construct produced using a CSDV nuclei acid molecule in order to generate transgenic plant resistant to CSDV. A 761pb nucleic acid fragment of the CSDV RNA-dependent RNA Polymerase of SEQ ID NO.: 1, were PCR amplified using the primers TY-Fw (5'-TG-GAGCTCCCTGCCAACGACCAAC-3') (SEQ ID NO: 6), containing a Sac I restriction site and TY-Rv (5'-TCTA-GAGCCTGGGGATGGAGAGC-3') (SEQ ID NO: 7), containing a Xba I restriction site. This fragment was cloned in PGEM-T easy vector (Promega corp, Madison, Wis., USA) and confirmed by sequencing. The RNA-dependent RNA Polymerase fragment was removed from pGEM-T by restriction digestion with Xba I and Sac I and cloned in the antisense direction in pA35S(2x) (Kay, R., Chan, A., Daly, M. & McPherson, J., 1987, *Science*, 236: 1299-1302), restricted with the same enzymes yielding the plasmid pATYMO-AS. The pA35S(2x) plasmid includes a herbicide resistance gene (Christensen A H, Quail P H., 1996, *Transgenic Res.* 15: 213-218.) under the control of the CaMV35S promoter. FIG. 9 represents a drawing of the TDNA insertion fragment of pATYMO-AS.

In order to produce the transgenic citrus containing the plasmid pATYMO-AS, explants from 'in vitro' germinated *Rangpur lime* were used for transformation experiments with *Agrobacterium tumefaciens* strain EHA105 carrying the plasmid. The explants were inoculated with an overnight bacterial suspension for 20 min, blotted dry and plated onto MS medium containing 2,4D and acetoseringone. The co-culti-

vation was made in darkness for 3 days and then the explants were plated in MS medium containing BAP, PPT, cefotaxime and vancomycin. The developed shoots were transferred to MS medium containing IBA, PPT, cefotaxime and vancomycin. Transgenic regenerated plants were tested by PCR and Southern blot.

For the southern blot experiment genomic DNA from transgenic and non-transgenic plants were isolated according to Dellaporta et al. (Dellaporta S. L., Wood J., Hicks J. B, 1983, *Plant Mol. Biol. Rep.*, 1:19-21.). Purified DNA was digested with EcoRI, separated by agarose gel electrophoresis (10 ug DNA/lane, 1% agarose) then transferred onto charged nylon membranes according to standard procedures (Sambrook et al., 2001 Molecular Cloning: A Laboratory Manual, 3rd ed., vol. 1-3, ed. Cold Spring Harbor Laboratory Press, Cold Spring Harbor, N.Y.). Probes were labeled using the Genes Images™ AlkPhos Direct™ labelling and detection system from Amersham Biosciences that is based on a dioxetane chemiluminescence system. Blots were hybridized to CSDV specific probes and the citrus chalcone synthase CitCHS2 gene (GenBank 5106368) as a positive control. Membranes were washed at 55° C. according to the instruction manual (Genes Images AlkPhos Direct labeling and detection system—Amersham Biosciences). Chemiluminescent signals were detected using a CDP-Star™ chemiluminescent detection reagent (Amersham Biosciences) according to the protocol recommended.

TABLE 1

RESULTS OF TEST FOR THE PRESENCE OF CSDV IN INSECTS COLLECTED FROM CITRUS THREES IN CSD AFFECTED AND NON-AFFECTED AREAS.			
	INSECT Leafhoppers/planthoppers	Samples Analyzed <sup>a</sup>	Diagnosis for CSDV
30	Group I	1 (affected area)	Negative
		2 (affected area)	Negative
35	Group II	1 (affected area)	Negative
		2 (affected area)	Negative
40	Group III	1 (affected area)	Negative
		2 (affected area)	Negative
	Group IV	1 (affected area)	Negative
	Group V	1 (affected area)	Negative
	Group VI	1 (affected area)	Negative
45	<i>T. citricida</i>	1 (non-affected area)	Negative
		2 (non-affected area)	Negative
		3 (non-affected area)	Negative
		4 (non-affected area)	Negative
50		5 (affected area)	Positive
		6 (affected area)	Positive
		7 (affected area)	Positive
		8 (affected area)	Positive
		9 (affected area)	Positive
55		10 (affected area)	Positive
		11 (affected area)	Positive
		12 (affected area)	Negative
		13 (affected area)	Positive
		14 (affected area)	Positive
60		15 (affected area)	Positive
	<i>A. spiraecola</i>	6 (non-affected area)	Negative
		7 (affected area)	Positive
		8 (affected area)	Positive
		9 (affected area)	Negative
65		10 (affected area)	Negative
	<i>A. gossypii</i>	5 (non-affected area)	Negative
		6 (non-affected area)	Negative
		7 (affected area)	Positive
		8 (affected area)	Negative
		9 (affected area)	Negative
		10 (affected area)	Negative

<sup>a</sup>each sample was composed of >50 aphids or around 10 leafhoppers/planthoppers.

## SEQUENCE LISTING

&lt;160&gt; NUMBER OF SEQ ID NOS: 19

<210> SEQ ID NO 1  
<211> LENGTH: 6821  
<212> TYPE: DNA  
<213> ORGANISM: Citrus Sudden Death Virus  
<220> FEATURE:  
<221> NAME/KEY: CDS  
<222> LOCATION: (110)..(6673)

&lt;400&gt; SEQUENCE: 1

gtccccctgtg atcgctctc ccgcgcctcca gccggaaaaga tatttttgc ttaactttc	60
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Met Asp Arg	
1	
atc tct gcc cgc att ccc gtc gcg ccc gct tcc gcc ggc ccg acc gag	166
Ile Ser Ala Arg Ile Pro Val Ala Pro Ala Ser Ala Gly Pro Thr Glu	
5 10 15	
tac act cca tac cca cac act cac cca ctc cta ccc cga ggt gtc ttc	214
Tyr Thr Pro Tyr Pro His Thr His Pro Leu Leu Pro Arg Gly Val Phe	
20 25 30 35	
acc tcc ggg cct att caa ccc tgt ctc cac ttt ctt cct cac cat gcc	262
Thr Ser Gly Pro Ile Gln Pro Cys Leu His Phe Leu Pro His His Ala	
40 45 50	
caa gat gcc ccc atc cgc tgc tac aga ccc ctc acc ttc gcc aac cat	310
Gln Asp Ala Pro Ile Arg Cys Tyr Arg Pro Leu Thr Phe Ala Asn His	
55 60 65	
ctc cgc tat gac cgt tcc gcc tca tcg ctc aag act ccc gtc aaa	358
Leu Arg Tyr Asp Arg Ser Ala Ser Ser Leu Lys Thr Pro Pro Val Lys	
70 75 80	
ctc cca ctg acc ggt ggt acc ctt gcc gat gcc atc ctt tcc ttg gca	406
Leu Pro Leu Thr Gly Gly Thr Leu Ala Asp Ala Ile Leu Ser Leu Ala	
85 90 95	
ccc acc act cac cgc gac acc atc gcc acc ccc ctc atg gaa gcc ctt	454
Pro Thr Thr His Arg Asp Thr Ile Ala Thr Pro Leu Met Glu Ala Leu	
100 105 110 115	
gct gaa cct tac cgc caa tcc ttg agc acc tac cca tgg cac att cca	502
Ala Glu Pro Tyr Arg Gln Ser Leu Ser Thr Tyr Pro Trp His Ile Pro	
120 125 130	
acc aat ctt cag ccc ttc ctc acc tct tgc gga atc acc act gct ggc	550
Thr Asn Leu Gln Pro Phe Leu Thr Ser Cys Gly Ile Thr Thr Ala Gly	
135 140 145	
caa ggc ttc aag gcc cac cct cac cca gtg cac aag acc atc gag acc	598
Gln Gly Phe Lys Ala His Pro His Pro Val His Lys Thr Ile Glu Thr	
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aat ctc ctc act aat gtc tgg ccc cac tac gcc acc act cct agt ggc	646
Asn Leu Leu Thr Asn Val Trp Pro His Tyr Ala Thr Thr Pro Ser Gly	
165 170 175	
gtc atg ttc atg aaa cca tca aag ttt gag aag ctc aaa atc aaa cag	694
Val Met Phe Met Lys Pro Ser Lys Phe Glu Lys Leu Lys Ile Lys Gln	
180 185 190 195	
ccc aac ttc tcc aag ctc tac aac tac cgc atc aca gcc aag gac acc	742
Pro Asn Phe Ser Lys Leu Tyr Asn Tyr Arg Ile Thr Ala Lys Asp Thr	
200 205 210	
acc cgt tac ccc tcc act tcc cca gac ttg ccc acc gag gac acc tgc	790
Thr Arg Tyr Pro Ser Thr Ser Pro Asp Leu Pro Thr Glu Asp Thr Cys	
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Phe Met His Asp Ala Leu Met Tyr Tyr Ser Pro Gly Gln Ile Cys Asp	
230 235 240	

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ctc ttc ctc tcc cgc ccc agc ctc caa aag ctc tat gct tcc ctt gtt Leu Phe Leu Ser Arg Pro Ser Leu Gln Lys Leu Tyr Ala Ser Leu Val 245 250 255	886
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tac cgc tac ccg atc gag aaa gat cag ctc atc tac gag ctc gag cag Tyr Arg Tyr Arg Glu Lys Asp Gln Leu Ile Tyr Glu Leu Glu Gln 280 285 290	982
aac ccc gcc cac aac tac atc cag cct cgc tct gcc atc gac tgg ctc Asn Pro Ala His Asn Tyr Ile Gln Pro Arg Ser Ala Ile Asp Trp Leu 295 300 305	1030
aag acc acc acc atc cgc tgc cag gac ctc act ctc acc atc tcc cgc Lys Thr Thr Ile Arg Cys Gln Asp Leu Thr Leu Thr Ile Ser Arg 310 315 320	1078
cta gat tcc tgg ggc cca gtc cac tct ctc ctg atc caa aga ggc aag Leu Asp Ser Trp Gly Pro Val His Ser Leu Leu Ile Gln Arg Gly Lys 325 330 335	1126
ccc cct atc cat ctt gag gag gac tcc atc agg ttc cgt gcc cca aaa Pro Pro Ile His Leu Glu Glu Asp Ser Ile Ser Phe Arg Ala Pro Lys 340 345 350 355	1174
gca gtc ctc ctg cct gag cca gct tca ctc tcc caa tca gtc cgc gac Ala Val Leu Leu Pro Glu Pro Ala Ser Leu Ser Gln Ser Val Arg Asp 360 365 370	1222
cgc ctg gtc cct gct gat gtt tac cag gct ctc ttc atc tat gtc cgg Arg Leu Val Pro Ala Asp Val Tyr Gln Ala Leu Phe Ile Tyr Val Arg 375 380 385	1270
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aat ctg gcc cac ttt gcc ttg gcc aca gct ccg cac aga ccc cac acc Asn Leu Ala His Phe Ala Leu Ala Thr Ala Pro His Arg Pro His Thr 420 425 430 435	1414
acc tac ttc ctg ttc aac tca acc gct gct cgg gtg gcc cat tgg ttc Thr Tyr Phe Leu Phe Asn Ser Thr Ala Ala Arg Val Ala His Trp Phe 440 445 450	1462
cgc act cat acc ctg gct ccg ctc tct gcc act gct gcc gcc gcg Arg Thr His Thr Leu Ala Pro Leu Ser Gly Ala Thr Ala Ala Ala 455 460 465	1510
agc ctt ctc atg acc gcc agc tgg gga ttc cgt gcc atg atc tcc tct Ser Leu Leu Met Thr Ala Ser Trp Gly Phe Arg Ala Met Ile Ser Ser 470 475 480	1558
cat ctt gtc tcc ctc tcc atc tgc aag cgc tgg ctc aaa gct cct cct His Leu Val Ser Leu Ser Ile Cys Lys Arg Trp Leu Lys Ala Pro Pro 485 490 495	1606
cat ctc ctc tgg ccc gag aaa gct ccc tgg ttc cag ctc acc ctg agg His Leu Leu Trp Pro Glu Lys Ala Pro Trp Phe Gln Leu Thr Leu Arg 500 505 510 515	1654
ccc aaa gtc act ggc cct ctg att gac ctg ccc att ctc cga ccc ttt Pro Lys Val Thr Gly Pro Leu Ile Asp Leu Pro Ile Leu Arg Pro Phe 520 525 530	1702
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Lys Phe Ile Gly Pro Asp Ser Pro Gln Asp Met His Asp Ser Tyr His		
580 585 590 595		
gcc atg ttt cat cca cag cct tgg ggc ctc act ctc act cgc aag gct		1942
Ala Met Phe His Pro Gln Pro Trp Gly Leu Thr Leu Thr Arg Lys Ala		
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615 620 625		
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645 650 655		
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Pro Gln Ser Ala Ser Ser Thr Gly Pro Ala Ser Asp Ser Arg Arg Ala		
660 665 670 675		
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Pro Gln Pro Ala Ser Ser Thr Gly Pro Asp Pro Pro Thr Gln Asn Thr		
680 685 690		
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Ser Ala Ala Pro Gln Pro Pro Ile Glu Ser Lys Val Thr Phe Ala Gln		
695 700 705		
ccc att gag agt gtg gca cct gta gtt cca gga gca gga gaa cct ccg		2278
Pro Ile Glu Ser Val Ala Pro Val Val Pro Gly Ala Gly Glu Pro Pro		
710 715 720		
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Gln Ser Ala Ser Ser Thr Gly Pro Ala Ser Val Ser Arg Arg Asp Pro		
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Gln Val Ala Ser Ser Thr Thr Pro Asp Ala Pro Thr Leu Asp Val Ser		
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Val Thr Pro Pro Lys Thr Ile Tyr Pro Ile Asp His Leu Gln Asn Asp		
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Phe Gly Pro Cys Arg Cys Ser Val Cys Glu Pro Leu Gln Pro Ala Pro		
775 780 785		
gtc ccc tcc act cct ctc acc gtc tcg gat cat aaa gaa gcc cag gac		2518
Val Pro Ser Thr Pro Leu Thr Val Ser Asp His Lys Glu Ala Gln Asp		
790 795 800		
gcc gaa gct ctt tcc tcg gcc ctc caa gcc ctc ggg ctc gct ccc acc		2566
Ala Glu Ala Leu Ser Ser Ala Leu Gln Ala Leu Gly Leu Ala Pro Thr		
805 810 815		
cca cca gct cca cag tct cag aac ctc act gta gag tcc tca gga gcc		2614
Pro Pro Ala Pro Gln Ser Gln Asn Leu Thr Val Glu Ser Ser Gly Ala		
820 825 830 835		
atg cat gcc tca tct tgg gat cag ctc tcc cca tca tct gac tgg		2662
Met His Ala Ser Ser Trp Asp Gln Leu Ser Ser Pro Ser Ser Asp Trp		
840 845 850		
gat cct tcc cct ctg gcc cgt gat agc tcc gcc tct ggt ccc cca ggc		2710
Asp Pro Ser Pro Leu Ala Arg Asp Ser Ser Ala Ser Gly Pro Pro Gly		
855 860 865		
atg tac tca gat ctc ttt cca gct ccc tac ctt cca ggc acc ggt cag		2758
Met Tyr Ser Asp Leu Phe Pro Ala Pro Tyr Leu Pro Gly Thr Gly Gln		

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870	875	880	
ttc atc ttc cgc tcc agg gcc aat ggt cggtt ccc aac atc cct tat ccc Phe Ile Phe Arg Ser Arg Ala Asn Gly Arg Ala Asn Ile Pro Tyr Pro 885 890 895			2806
gac atg gat tgc ctc ttg ctt tcc atc gag caa gcc acc cgc ctt ccc Asp Met Asp Cys Leu Leu Ser Ile Glu Gln Ala Thr Arg Leu Pro 900 905 910 915			2854
aag gag gct ctc tgg gac acc ctc tgt gcc aca tgc ccc gac tct ctc Lys Glu Ala Leu Trp Asp Thr Leu Cys Ala Thr Cys Pro Asp Ser Leu 920 925 930			2902
ctt gat cct gat acc att cgc cga gtc gga ttg tcc act gac cac ttt Leu Asp Pro Asp Thr Ile Arg Arg Val Gly Leu Ser Thr Asp His Phe 935 940 945			2950
gcc atc ctg gcc cac cac tac tcc ctc agg tgc cgc ttt cac acc gcc Ala Ile Leu Ala His His Tyr Ser Leu Arg Cys Arg Phe His Thr Ala 950 955 960			2998
cat ggt gtc att gag ctc ggc atg gct gat gcc acc tcc tca ttc gac His Gly Val Ile Glu Leu Gly Met Ala Asp Ala Thr Ser Ser Phe Asp 965 970 975			3046
atc gac cac act gct ggc aac ccc ggc cac ttc tcc ctc cgg caa tct Ile Asp His Thr Ala Gly Asn Pro Gly His Phe Ser Leu Arg Gln Ser 980 985 990 995			3094
gcc act ccg agg cta aat gga gga att gct caa gat ctc gct gtg Ala Thr Pro Arg Leu Asn Gly Gly Ile Ala Gln Asp Leu Ala Val 1000 1005 1010			3139
gcc gct ctc agg ttc aac att gat ggc act ctc ctc cca atc cgc Ala Ala Leu Arg Phe Asn Ile Asp Gly Thr Leu Leu Pro Ile Arg 1015 1020 1025			3184
tca gtt cat gtc tat tcc act tgg cca aag aga gca aag aac ctg Ser Val His Val Tyr Ser Thr Trp Pro Lys Arg Ala Lys Asn Leu 1030 1035 1040			3229
tcg tcg aac atg aag aac ggc ttt gac ggc atc atg gcc aac atc Ser Ser Asn Met Lys Asn Gly Phe Asp Gly Ile Met Ala Asn Ile 1045 1050 1055			3274
cac ccc acc aag acc aat gaa tcg aga gag aag atc ttg gca ctc His Pro Thr Lys Thr Asn Glu Ser Arg Glu Lys Ile Leu Ala Leu 1060 1065 1070			3319
gat tcg cag ctg gac atc gct gtc agg aga tcc gtc cgt ctg atc Asp Ser Gln Leu Asp Ile Ala Val Arg Arg Ser Val Arg Leu Ile 1075 1080 1085			3364
cat att gcc ggg ttc cca ggg tgc ggc aag tcc ttt ccc atc tcc His Ile Ala Gly Phe Pro Gly Cys Gly Lys Ser Phe Pro Ile Ser 1090 1095 1100			3409
cgc ctc ctc cgc act cca acc ttc agg aac ttt aag gtg gca gtt Arg Leu Leu Arg Thr Pro Thr Phe Arg Asn Phe Lys Val Ala Val 1105 1110 1115			3454
ccc act gtt gag ctc cga gcc gag tgg aaa acc att act ggt ctc Pro Thr Val Glu Leu Arg Ala Glu Trp Lys Thr Ile Thr Gly Leu 1120 1125 1130			3499
ccg gcc tca gaa gcc tgg cgc atc ggc acc tgg gaa tcc tct ctc Pro Ala Ser Glu Ala Trp Arg Ile Gly Thr Trp Glu Ser Ser Leu 1135 1140 1145			3544
ctc aag tct gcc cgg gtc ctg gtc att gat gaa atc tac aag atg Leu Lys Ser Ala Arg Val Leu Val Ile Asp Glu Ile Tyr Lys Met 1150 1155 1160			3589
cca aga ggc tac att gat ctc gcc atc cac tct gat ccc acc att Pro Arg Gly Tyr Ile Asp Leu Ala Ile His Ser Asp Pro Thr Ile 1165 1170 1175			3634
gaa atg gtc att gct ctc ggt gat cca ctc caa gga gag tac cac			3679

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Glu Met Val Ile Ala	Leu Gly Asp Pro Leu	Gln Gly Glu Tyr His	
1180	1185	1190	
tcc act cat cct tcc	tct acc aac tcc cgc	ctt ctc tct gag ccc	3724
Ser Thr His Pro Ser	Ser Thr Asn Ser Arg	Leu Leu Ser Glu Pro	
1195	1200	1205	
cag cat ctc tcc atg	tac ctt gac ttc tac	tgc ttg tgg tcc cac	3769
Gln His Leu Ser Met	Tyr Leu Asp Phe Tyr	Cys Leu Trp Ser His	
1210	1215	1220	
cgc gtt ccg cag aac	gtg gcc gcc ttc ttc	cat gtc aag acc acc	3814
Arg Val Pro Gln Asn	Val Ala Ala Phe Phe	His Val Lys Thr Thr	
1225	1230	1235	
tcc aaa cag cct ggc	ttc tgc cgc tac cag	aga gag ctg ccg aac	3859
Ser Lys Gln Pro Gly	Phe Cys Arg Tyr Gln	Arg Glu Leu Pro Asn	
1240	1245	1250	
tcc aga atc ctg gcc	aac tct cag aat gca	ggc cat acc ctc cag	3904
Ser Arg Ile Leu Ala	Asn Ser Gln Asn Ala	Gly His Thr Leu Gln	
1255	1260	1265	
cag tgt ggc tac gct	gcc gtc acc att gcc	tcc agt cag ggc tcc	3949
Gln Cys Gly Tyr Ala	Ala Val Thr Ile Ala	Ser Ser Gln Gly Ser	
1270	1275	1280	
acc tat gaa aat gcg	gcc tgc att cac ctg	gac cga aac agc tcc	3994
Thr Tyr Glu Asn Ala	Ala Cys Ile His Leu	Asp Arg Asn Ser Ser	
1285	1290	1295	
ttg ctc tcc cct gct	cac tcc atg gtt gct	ctc act cgc tca aag	4039
Leu Leu Ser Pro Ala	His Ser Met Val Ala	Leu Thr Arg Ser Lys	
1300	1305	1310	
gtt ggt gtc atc ttc	acc ggc gat ccc gcc	cag ctc tcc aat gct	4084
Val Gly Val Ile Phe	Thr Gly Asp Pro Ala	Gln Leu Ser Asn Ala	
1315	1320	1325	
cca agc tcc aac cga	atg ttc tca gag ttc	ttc tca ggc cgc acc	4129
Pro Ser Ser Asn Arg	Met Phe Ser Glu Phe	Phe Ser Gly Arg Thr	
1330	1335	1340	
cgc cct ctt cat gac	tgg ttc cac aat gag	ttc cca aag gcc act	4174
Arg Pro Leu His Asp	Trp Phe His Asn Glu	Phe Pro Lys Ala Thr	
1345	1350	1355	
gtc ctc acc gag ccc	ctc aag act cgg ggg	ccc cgc ctc acc ggt	4219
Val Leu Thr Glu Pro	Leu Lys Thr Arg Gly	Pro Arg Leu Thr Gly	
1360	1365	1370	
gct gcc tca cca tac	tcc aag gct gtc cca	atc cgc caa gcc tcc	4264
Ala Ala Ser Pro Tyr	Ser Lys Ala Val Pro	Ile Arg Gln Ala Ser	
1375	1380	1385	
acc cca gct ctc aag	cct gat ttc caa ggg	gac gtc ata atc tca	4309
Thr Pro Ala Leu Lys	Pro Asp Phe Gln Gly	Asp Val Ile Ile Ser	
1390	1395	1400	
gca ccc ata gtt ctc	ggc tcc ggc gag ctc	aat gcc cct caa gtc	4354
Ala Pro Ile Val Leu	Gly Ser Gly Glu Leu	Asn Ala Pro Gln Val	
1405	1410	1415	
tcc tct cac ttc ctc	ccc gag act cgc cgt	cct ctc cac tgg gac	4399
Ser Ser His Phe Leu	Pro Glu Thr Arg Arg	Pro Leu His Trp Asp	
1420	1425	1430	
att cca tct gcc atc	cct gag agt gcc acc	aga ccg gac tcc act	4444
Ile Pro Ser Ala Ile	Pro Glu Ser Ala Thr	Arg Pro Asp Ser Thr	
1435	1440	1445	
gag ccc acc acc tcc	cat cca gag cca gtc	tac ccc ggg gaa act	4489
Glu Pro Thr Thr Ser	His Pro Glu Pro Val	Tyr Pro Gly Glu Thr	
1450	1455	1460	
ttt gag aat ctt gct	gcc cac ttt ctc cct	gcc cac gac cca acc	4534
Phe Glu Asn Leu Ala	Ala His Phe Leu Pro	Ala His Asp Pro Thr	
1465	1470	1475	

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gat cgt gag atc tac	tgg cag ggt cag ctg	tcc aac cag ttc cca	4579
Asp Arg Glu Ile Tyr	Trp Gln Gly Gln Leu	Ser Asn Gln Phe Pro	
1480	1485	1490	
cac atg gac aag gaa	ttc cat ttg gct gca	caa ccc atg agt ctc	4624
His Met Asp Lys Glu	Phe His Leu Ala Ala	Gln Pro Met Ser Leu	
1495	1500	1505	
ctg gct gcc gtt cat	caa gag aag caa gat	ccc act cta ctg cca	4669
Leu Ala Ala Val His	Gln Glu Lys Gln Asp	Pro Thr Leu Leu Pro	
1510	1515	1520	
gct tca atc caa aag	aga ctc cgc ttc cgc	ccc tcc gac aag ccc	4714
Ala Ser Ile Gln Lys	Arg Leu Arg Phe Arg	Pro Ser Asp Lys Pro	
1525	1530	1535	
tac cag atc acc cca	aaa gat gaa atc ctg	ggc cag ctc ctc ttt	4759
Tyr Gln Ile Thr Pro	Lys Asp Glu Ile Leu	Gly Gln Leu Leu Phe	
1540	1545	1550	
gaa ggc ctc tgc cga	gcc tac cac aga tct	cca ttt cac act gag	4804
Glu Gly Leu Cys Arg	Ala Tyr His Arg Ser	Pro Phe His Thr Glu	
1555	1560	1565	
gcc ttt gat ccc gtg	ctt ttc gcc gag tgc	atc aat ctc aat gag	4849
Ala Phe Asp Pro Val	Leu Phe Ala Glu Cys	Ile Asn Leu Asn Glu	
1570	1575	1580	
ttc gcc cag ctc tcg	tcc aag acc cag gct	act att atg ggc aat	4894
Phe Ala Gln Leu Ser	Ser Lys Thr Gln Ala	Thr Ile Met Gly Asn	
1585	1590	1595	
gct cgc cgc tca gac	cct gat tgg cgg tgg	agc gca gtt cgc atc	4939
Ala Arg Arg Ser Asp	Pro Asp Trp Arg Trp	Ser Ala Val Arg Ile	
1600	1605	1610	
ttc tcc aag acc caa	cac aag gtg aat gaa	ggg tcc att ttc cgc	4984
Phe Ser Lys Thr Gln	His Lys Val Asn Glu	Gly Ser Ile Phe Arg	
1615	1620	1625	
tcc tgg aag gcc tgc	caa act ttg gct ctc	atg cat gat gct gtt	5029
Ser Trp Lys Ala Cys	Gln Thr Leu Ala Leu	Met His Asp Ala Val	
1630	1635	1640	
gtt cta atc ctg ggc	cct gtc aag aag tac	cag cga gtc ttt gat	5074
Val Leu Ile Leu Gly	Pro Val Lys Lys Tyr	Gln Arg Val Phe Asp	
1645	1650	1655	
cag aga gac cga ccc	cga cac ctt tac atc	cat gca ggc aac act	5119
Gln Arg Asp Arg Pro	Arg His Leu Tyr Ile	His Ala Gly Asn Thr	
1660	1665	1670	
cca tca caa atg agc	aac tgg tgt caa cag	cat ctc act act gcc	5164
Pro Ser Gln Met Ser	Asn Trp Cys Gln Gln	His Leu Thr Thr Ala	
1675	1680	1685	
gtc aag ttg gcc aat	gac tac act gcc ttc	gac cag tct cag cat	5209
Val Lys Leu Ala Asn	Asp Tyr Thr Ala Phe	Asp Gln Ser Gln His	
1690	1695	1700	
ggt gag gcg gtc gtc	ctt gaa aga aag aaa	atg gaa aga ctc tcc	5254
Gly Glu Ala Val Val	Leu Glu Arg Lys Lys	Met Glu Arg Leu Ser	
1705	1710	1715	
atc ccc cag gct ctc	att gat ctt cac atc	cat ctc aaa acc cat	5299
Ile Pro Gln Ala Leu	Ile Asp Leu His Ile	His Leu Lys Thr His	
1720	1725	1730	
gtt tcc acc cag ttt	ggc ccc ctc aca tgc	atg cgc ctg act ggc	5344
Val Ser Thr Gln Phe	Gly Pro Leu Thr Cys	Met Arg Leu Thr Gly	
1735	1740	1745	
gag cct ggc act tat	gat gat aac tct gac	tac aat ctt gca gtt	5389
Glu Pro Gly Thr Tyr	Asp Asp Asn Ser Asp	Tyr Asn Leu Ala Val	
1750	1755	1760	
gtc aac tgt gag tac	atg gct gcc aac act	ccc act atg gtc tca	5434
Val Asn Cys Glu Tyr	Met Ala Ala Asn Thr	Pro Thr Met Val Ser	
1765	1770	1775	

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ggc gac gac tcc ctc	ctg gat cgt gag cct	ccc act cgc cct gaa	5479
Gly Asp Asp Ser Leu	Leu Asp Arg Glu Pro	Pro Thr Arg Pro Glu	
1780	1785	1790	
tgg gtc atc ctc cag	cct ctt ctc agt ctc	cgc ttc aag aaa gaa	5524
Trp Val Ile Leu Gln	Pro Leu Leu Ser Leu	Arg Phe Lys Lys Glu	
1795	1800	1805	
agg ggt cgg tac gcc	acc ttc tgt ggc tac	tac gcc tcc cat gtc	5569
Arg Gly Arg Tyr Ala	Thr Phe Cys Gly Tyr	Tyr Ala Ser His Val	
1810	1815	1820	
ggc tgt gtc cgc tcc	ccc gtg gct ctc ttt	gcc aag ctg gcc ata	5614
Gly Cys Val Arg Ser	Pro Val Ala Leu Phe	Ala Lys Leu Ala Ile	
1825	1830	1835	
gtc gtc gat gac ggc	tcc atc tct gac aaa	atg gcc tca tac ctc	5659
Ala Val Asp Asp Gly	Ser Ile Ser Asp Lys	Met Ala Ser Tyr Leu	
1840	1845	1850	
tct gaa ttt gct ctt	ggc cac tcc ctt gga	gac cat ctc tgg gaa	5704
Ser Glu Phe Ala Leu	Gly His Ser Leu Gly	Asp His Leu Trp Glu	
1855	1860	1865	
gtc ttg ccc ctc gag	gcc gtt ccc ttc caa	tct gcc tgc ttt gac	5749
Ala Leu Pro Leu Glu	Ala Val Pro Phe Gln	Ser Ala Cys Phe Asp	
1870	1875	1880	
ttc ttc tgc cgc cgg	gcc ccc aga cac ctc	aaa ctc tct ctc atg	5794
Phe Phe Cys Arg Arg	Ala Pro Arg His Leu	Lys Leu Ser Leu Met	
1885	1890	1895	
ctc ggc gag gtc cca	gaa tcc atc att gcc	cgc atc ggg tca tcc	5839
Leu Gly Glu Val Pro	Glu Ser Ile Ile Ala	Arg Ile Gly Ser Ser	
1900	1905	1910	
ttg aag tgg gcc tct	cat gcc atc tac acc	aca ctc tcc tct gcc	5884
Leu Lys Trp Ala Ser	His Ala Ile Tyr Thr	Thr Leu Ser Ser Ala	
1915	1920	1925	
gct cga gtg gcc att	ctg aga tcc tcc cgc	aac agc aga tcc atg	5929
Ala Arg Val Ala Ile	Leu Arg Ser Ser Arg	Asn Ser Arg Ser Met	
1930	1935	1940	
cca gat gac ccc gag	acc act ctg cta caa	ggg gaa ttg ctt cag	5974
Pro Asp Asp Pro Asp	Thr Thr Leu Leu Gln	Gly Glu Leu Leu Gln	
1945	1950	1955	
cac ttt caa gta cca	ttc atg caa tct gac	act ctc ctg cct ctc	6019
His Phe Gln Val Pro	Phe Met Gln Ser Asp	Thr Leu Leu Pro Leu	
1960	1965	1970	
act ggt ggt tcc tct	gct ccc atc ctc aca	cca gaa gcc ttc tcc	6064
Thr Gly Gly Ser Ser	Ala Pro Ile Leu Thr	Pro Glu Ala Phe Ser	
1975	1980	1985	
acc tcc ctc gcc ttc	tcc atg gcc agc gat	gcc caa gca ggt ccg	6109
Thr Ser Leu Ala Phe	Ser Met Ala Ser Asp	Ala Gln Ala Gly Pro	
1990	1995	2000	
gcc ccc agt cgc gat	gat cgc gtt gac cgc	cag cct cgc ctt cct	6154
Ala Pro Ser Arg Asp	Asp Arg Val Asp Arg	Gln Pro Arg Leu Pro	
2005	2010	2015	
gct gct cct cgc gtt	gct gaa gtt ggt ctc	aat gcc ccg tcg gtc	6199
Ala Ala Pro Arg Val	Ala Glu Val Gly Leu	Asn Ala Pro Ser Val	
2020	2025	2030	
gac tac ccg ttc cag	tgg gtc gtc gcc tcc	tac gac gga tca gaa	6244
Asp Tyr Pro Phe Gln	Trp Val Val Ala Ser	Tyr Asp Gly Ser Glu	
2035	2040	2045	
gcc aag aac cta agt	gat gat ctc tct ggc	tct gcc act ctc acc	6289
Ala Lys Asn Leu Ser	Asp Asp Leu Ser Gly	Ser Ala Thr Leu Thr	
2050	2055	2060	
aaa gtc atg gcc aac	tac cga cat gct gag	ctc aca tct gtt gag	6334
Lys Val Met Ala Asn	Tyr Arg His Ala Glu	Leu Thr Ser Val Glu	

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2065	2070	2075	
ctg gag gtc tgc cct	ctt gct gca gcc ttc	tcc aag ccc atc tct	6379
Leu Glu Val Cys Pro	Leu Ala Ala Ala Phe	Ser Lys Pro Ile Ser	
2080	2085	2090	
gtg tcg gcc gtc tgg	acc att gcc tcc atc	tct cca gct tcc gcc	6424
Val Ser Ala Val Trp	Thr Ile Ala Ser Ile	Ser Pro Ala Ser Ala	
2095	2100	2105	
tct gaa acc tcc tac	tat ggc ggt cga ctc	ttc act gtt ggc ggt	6469
Ser Glu Thr Ser Tyr	Tyr Gly Gly Arg Leu	Phe Thr Val Gly Gly	
2110	2115	2120	
cct gtc ctc atg tcc	agc acc acc cat ctc	cct gct gat ctc acc	6514
Pro Val Leu Met Ser	Ser Thr Thr His Leu	Pro Ala Asp Leu Thr	
2125	2130	2135	
cgc ctc aat cct gtg	ctc aag ggc ccc gtc	aag tac aca gac tgc	6559
Arg Leu Asn Pro Val	Leu Lys Gly Pro Val	Lys Tyr Thr Asp Cys	
2140	2145	2150	
ccc aga ttc tcc tac	tcc gtc tac tcc aat	ggc gga acc aag ggc	6604
Pro Arg Phe Ser Tyr	Ser Val Tyr Ser Asn	Gly Gly Thr Lys Gly	
2155	2160	2165	
acc aat ctc tgc acc	atc atc ctc cgg gga	gtt gtc cgc ctc agc	6649
Thr Asn Leu Cys Thr	Ile Ile Leu Arg Gly	Val Val Arg Leu Ser	
2170	2175	2180	
ggc ccc tcc ggt aat	ctt ctc gct taggcgagcc	tcttcagggt aaggaaaaca	6703
Gly Pro Ser Gly Asn	Leu Leu Ala		
2185			
cctcctggtc tcagccaggt	aatgatgcta aacctcccc	gctcaagcag caatgcctag	6763
ggttgccggt cgatccaaag	accgttttc ttattttttt	aataaaaaaa aaaaaaaaa	6821
<210> SEQ_ID NO 2			
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Met Asp Arg Ile Ser Ala Arg Ile Pro Val Ala Pro Ala Ser Ala Gly			
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Pro Thr Glu Tyr Thr Pro Tyr Pro His Thr His Pro Leu Leu Pro Arg			
20 25 30			
Gly Val Phe Thr Ser Gly Pro Ile Gln Pro Cys Leu His Phe Leu Pro			
35 40 45			
His His Ala Gln Asp Ala Pro Ile Arg Cys Tyr Arg Pro Leu Thr Phe			
50 55 60			
Ala Asn His Leu Arg Tyr Asp Arg Ser Ala Ser Ser Leu Lys Thr Pro			
65 70 75 80			
Pro Val Lys Leu Pro Leu Thr Gly Thr Leu Ala Asp Ala Ile Leu			
85 90 95			
Ser Leu Ala Pro Thr Thr His Arg Asp Thr Ile Ala Thr Pro Leu Met			
100 105 110			
Glu Ala Leu Ala Glu Pro Tyr Arg Gln Ser Leu Ser Thr Tyr Pro Trp			
115 120 125			
His Ile Pro Thr Asn Leu Gln Pro Phe Leu Thr Ser Cys Gly Ile Thr			
130 135 140			
Thr Ala Gly Gln Gly Phe Lys Ala His Pro His Pro Val His Lys Thr			
145 150 155 160			
Ile Glu Thr Asn Leu Leu Thr Asn Val Trp Pro His Tyr Ala Thr Thr			
165 170 175			

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Pro Ser Gly Val Met Phe Met Lys Pro Ser Lys Phe Glu Lys Leu Lys  
 180 185 190  
 Ile Lys Gln Pro Asn Phe Ser Lys Leu Tyr Asn Tyr Arg Ile Thr Ala  
 195 200 205  
 Lys Asp Thr Thr Arg Tyr Pro Ser Thr Ser Pro Asp Leu Pro Thr Glu  
 210 215 220  
 Asp Thr Cys Phe Met His Asp Ala Leu Met Tyr Tyr Ser Pro Gly Gln  
 225 230 235 240  
 Ile Cys Asp Leu Phe Leu Ser Arg Pro Ser Leu Gln Lys Leu Tyr Ala  
 245 250 255  
 Ser Leu Val Val Pro Pro Glu Ser Asp Phe Thr Thr Ile Ser Leu Phe  
 260 265 270  
 Pro Asp Leu Tyr Arg Tyr Arg Ile Glu Lys Asp Gln Leu Ile Tyr Glu  
 275 280 285  
 Leu Glu Gln Asn Pro Ala His Asn Tyr Ile Gln Pro Arg Ser Ala Ile  
 290 295 300  
 Asp Trp Leu Lys Thr Thr Thr Ile Arg Cys Gln Asp Leu Thr Leu Thr  
 305 310 315 320  
 Ile Ser Arg Leu Asp Ser Trp Gly Pro Val His Ser Leu Leu Ile Gln  
 325 330 335  
 Arg Gly Lys Pro Pro Ile His Leu Glu Glu Asp Ser Ile Ser Phe Arg  
 340 345 350  
 Ala Pro Lys Ala Val Leu Leu Pro Glu Pro Ala Ser Leu Ser Gln Ser  
 355 360 365  
 Val Arg Asp Arg Leu Val Pro Ala Asp Val Tyr Gln Ala Leu Phe Ile  
 370 375 380  
 Tyr Val Arg Ala Val Arg Thr Leu Arg Val Thr Asp Pro Ala Gly Phe  
 385 390 395 400  
 Val Arg Thr Gln Ile Ser Lys Pro Glu Tyr Ser Trp Val Thr Ser Ser  
 405 410 415  
 Ala Trp Asp Asn Leu Ala His Phe Ala Leu Ala Thr Ala Pro His Arg  
 420 425 430  
 Pro His Thr Thr Tyr Phe Leu Phe Asn Ser Thr Ala Ala Arg Val Ala  
 435 440 445  
 His Trp Phe Arg Thr His Thr Leu Ala Pro Leu Ser Gly Ala Thr Ala  
 450 455 460  
 Ala Ala Ala Ser Leu Leu Met Thr Ala Ser Trp Gly Phe Arg Ala Met  
 465 470 475 480  
 Ile Ser Ser His Leu Val Ser Leu Ser Ile Cys Lys Arg Trp Leu Lys  
 485 490 495  
 Ala Pro Pro His Leu Leu Trp Pro Glu Lys Ala Pro Trp Phe Gln Leu  
 500 505 510  
 Thr Leu Arg Pro Lys Val Thr Gly Pro Leu Ile Asp Leu Pro Ile Leu  
 515 520 525  
 Arg Pro Phe Arg Leu Phe Pro Ser Thr Cys Ala Lys Leu Gly Ala Lys  
 530 535 540  
 His Pro Ala Leu Ala Thr Leu Leu Pro Ala Ala Pro Arg Pro Thr Trp  
 545 550 555 560  
 Pro Leu Lys Val Gly Leu Ala Leu Ala Val Pro Val Cys Leu Phe  
 565 570 575  
 Leu Trp Arg Lys Phe Ile Gly Pro Asp Ser Pro Gln Asp Met His Asp  
 580 585 590  
 Ser Tyr His Ala Met Phe His Pro Gln Pro Trp Gly Leu Thr Leu Thr

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595	600	605
Arg Lys Ala Ile Cys Cys Asp Arg Ala Pro Phe Leu Pro Ile Pro Val		
610	615	620
Val Pro Ser Ser Asp Phe Lys Ala Pro Pro Thr Pro Ala Thr Pro Leu		
625	630	635
640		
Leu Thr Ser Ile Pro Ile Lys Gly Val Glu Pro Gln Val Ser Gly Glu		
645	650	655
Gly Val Pro Pro Gln Ser Ala Ser Ser Thr Gly Pro Ala Ser Asp Ser		
660	665	670
Arg Arg Ala Pro Gln Pro Ala Ser Ser Thr Gly Pro Asp Pro Pro Thr		
675	680	685
Gln Asn Thr Ser Ala Ala Pro Gln Pro Pro Ile Glu Ser Lys Val Thr		
690	695	700
Phe Ala Gln Pro Ile Glu Ser Val Ala Pro Val Val Pro Gly Ala Gly		
705	710	715
720		
Glu Pro Pro Gln Ser Ala Ser Ser Thr Gly Pro Ala Ser Val Ser Arg		
725	730	735
Arg Asp Pro Gln Val Ala Ser Ser Thr Thr Pro Asp Ala Pro Thr Leu		
740	745	750
Asp Val Ser Val Thr Pro Pro Lys Thr Ile Tyr Pro Ile Asp His Leu		
755	760	765
Gln Asn Asp Phe Gly Pro Cys Arg Cys Ser Val Cys Glu Pro Leu Gln		
770	775	780
Pro Ala Pro Val Pro Ser Thr Pro Leu Thr Val Ser Asp His Lys Glu		
785	790	795
800		
Ala Gln Asp Ala Glu Ala Leu Ser Ser Ala Leu Gln Ala Leu Gly Leu		
805	810	815
Ala Pro Thr Pro Pro Ala Pro Gln Ser Gln Asn Leu Thr Val Glu Ser		
820	825	830
Ser Gly Ala Met His Ala Ser Ser Trp Asp Gln Leu Ser Ser Pro Ser		
835	840	845
Ser Asp Trp Asp Pro Ser Pro Leu Ala Arg Asp Ser Ser Ala Ser Gly		
850	855	860
Pro Pro Gly Met Tyr Ser Asp Leu Phe Pro Ala Pro Tyr Leu Pro Gly		
865	870	875
880		
Thr Gly Gln Phe Ile Phe Arg Ser Arg Ala Asn Gly Arg Ala Asn Ile		
885	890	895
Pro Tyr Pro Asp Met Asp Cys Leu Leu Ser Ile Glu Gln Ala Thr		
900	905	910
Arg Leu Pro Lys Glu Ala Leu Trp Asp Thr Leu Cys Ala Thr Cys Pro		
915	920	925
Asp Ser Leu Leu Asp Pro Asp Thr Ile Arg Arg Val Gly Leu Ser Thr		
930	935	940
Asp His Phe Ala Ile Leu Ala His His Tyr Ser Leu Arg Cys Arg Phe		
945	950	955
960		
His Thr Ala His Gly Val Ile Glu Leu Gly Met Ala Asp Ala Thr Ser		
965	970	975
Ser Phe Asp Ile Asp His Thr Ala Gly Asn Pro Gly His Phe Ser Leu		
980	985	990
Arg Gln Ser Ala Thr Pro Arg Leu Asn Gly Gly Ile Ala Gln Asp Leu		
995	1000	1005
Ala Val Ala Ala Leu Arg Phe Asn Ile Asp Gly Thr Leu Leu Pro		
1010	1015	1020

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Ile Arg Ser Val His Val Tyr Ser Thr Trp Pro Lys Arg Ala Lys  
1025 1030 1035

Asn Leu Ser Ser Asn Met Lys Asn Gly Phe Asp Gly Ile Met Ala  
1040 1045 1050

Asn Ile His Pro Thr Lys Thr Asn Glu Ser Arg Glu Lys Ile Leu  
1055 1060 1065

Ala Leu Asp Ser Gln Leu Asp Ile Ala Val Arg Arg Ser Val Arg  
1070 1075 1080

Leu Ile His Ile Ala Gly Phe Pro Gly Cys Gly Lys Ser Phe Pro  
1085 1090 1095

Ile Ser Arg Leu Leu Arg Thr Pro Thr Phe Arg Asn Phe Lys Val  
1100 1105 1110

Ala Val Pro Thr Val Glu Leu Arg Ala Glu Trp Lys Thr Ile Thr  
1115 1120 1125

Gly Leu Pro Ala Ser Glu Ala Trp Arg Ile Gly Thr Trp Glu Ser  
1130 1135 1140

Ser Leu Leu Lys Ser Ala Arg Val Leu Val Ile Asp Glu Ile Tyr  
1145 1150 1155

Lys Met Pro Arg Gly Tyr Ile Asp Leu Ala Ile His Ser Asp Pro  
1160 1165 1170

Thr Ile Glu Met Val Ile Ala Leu Gly Asp Pro Leu Gln Gly Glu  
1175 1180 1185

Tyr His Ser Thr His Pro Ser Ser Thr Asn Ser Arg Leu Leu Ser  
1190 1195 1200

Glu Pro Gln His Leu Ser Met Tyr Leu Asp Phe Tyr Cys Leu Trp  
1205 1210 1215

Ser His Arg Val Pro Gln Asn Val Ala Ala Phe Phe His Val Lys  
1220 1225 1230

Thr Thr Ser Lys Gln Pro Gly Phe Cys Arg Tyr Gln Arg Glu Leu  
1235 1240 1245

Pro Asn Ser Arg Ile Leu Ala Asn Ser Gln Asn Ala Gly His Thr  
1250 1255 1260

Leu Gln Gln Cys Gly Tyr Ala Ala Val Thr Ile Ala Ser Ser Gln  
1265 1270 1275

Gly Ser Thr Tyr Glu Asn Ala Ala Cys Ile His Leu Asp Arg Asn  
1280 1285 1290

Ser Ser Leu Leu Ser Pro Ala His Ser Met Val Ala Leu Thr Arg  
1295 1300 1305

Ser Lys Val Gly Val Ile Phe Thr Gly Asp Pro Ala Gln Leu Ser  
1310 1315 1320

Asn Ala Pro Ser Ser Asn Arg Met Phe Ser Glu Phe Phe Ser Gly  
1325 1330 1335

Arg Thr Arg Pro Leu His Asp Trp Phe His Asn Glu Phe Pro Lys  
1340 1345 1350

Ala Thr Val Leu Thr Glu Pro Leu Lys Thr Arg Gly Pro Arg Leu  
1355 1360 1365

Thr Gly Ala Ala Ser Pro Tyr Ser Lys Ala Val Pro Ile Arg Gln  
1370 1375 1380

Ala Ser Thr Pro Ala Leu Lys Pro Asp Phe Gln Gly Asp Val Ile  
1385 1390 1395

Ile Ser Ala Pro Ile Val Leu Gly Ser Gly Glu Leu Asn Ala Pro  
1400 1405 1410

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Trp	Asp	Ile	Pro	Ser	Ala	Ile	Pro	Glu	Ser	Ala	Thr	Arg	Pro	Asp
1430					1435							1440		
Ser	Thr	Glu	Pro	Thr	Thr	Ser	His	Pro	Glu	Pro	Val	Tyr	Pro	Gly
1445					1450							1455		
Glu	Thr	Phe	Glu	Asn	Leu	Ala	Ala	His	Phe	Leu	Pro	Ala	His	Asp
1460					1465						1470			
Pro	Thr	Asp	Arg	Glu	Ile	Tyr	Trp	Gln	Gly	Gln	Leu	Ser	Asn	Gln
1475					1480						1485			
Phe	Pro	His	Met	Asp	Lys	Glu	Phe	His	Leu	Ala	Ala	Gln	Pro	Met
1490					1495						1500			
Ser	Leu	Leu	Ala	Ala	Val	His	Gln	Glu	Lys	Gln	Asp	Pro	Thr	Leu
1505					1510						1515			
Leu	Pro	Ala	Ser	Ile	Gln	Lys	Arg	Leu	Arg	Phe	Arg	Pro	Ser	Asp
1520					1525						1530			
Lys	Pro	Tyr	Gln	Ile	Thr	Pro	Lys	Asp	Glu	Ile	Leu	Gly	Gln	Leu
1535					1540						1545			
Leu	Phe	Glu	Gly	Leu	Cys	Arg	Ala	Tyr	His	Arg	Ser	Pro	Phe	His
1550					1555						1560			
Thr	Glu	Ala	Phe	Asp	Pro	Val	Leu	Phe	Ala	Glu	Cys	Ile	Asn	Leu
1565					1570						1575			
Asn	Glu	Phe	Ala	Gln	Leu	Ser	Ser	Lys	Thr	Gln	Ala	Thr	Ile	Met
1580					1585						1590			
Gly	Asn	Ala	Arg	Arg	Ser	Asp	Pro	Asp	Trp	Arg	Trp	Ser	Ala	Val
1595					1600						1605			
Arg	Ile	Phe	Ser	Lys	Thr	Gln	His	Lys	Val	Asn	Glu	Gly	Ser	Ile
1610					1615						1620			
Phe	Arg	Ser	Trp	Lys	Ala	Cys	Gln	Thr	Leu	Ala	Leu	Met	His	Asp
1625					1630						1635			
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1640					1645						1650			
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1655					1660						1665			
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1670					1675						1680			
Thr	Ala	Val	Lys	Leu	Ala	Asn	Asp	Tyr	Thr	Ala	Phe	Asp	Gln	Ser
1685					1690						1695			
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1700					1705						1710			
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1715					1720						1725			
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1730					1735						1740			
Thr	Gly	Glu	Pro	Gly	Thr	Tyr	Asp	Asp	Asn	Ser	Asp	Tyr	Asn	Leu
1745					1750						1755			
Ala	Val	Val	Asn	Cys	Glu	Tyr	Met	Ala	Ala	Asn	Thr	Pro	Thr	Met
1760					1765						1770			
Val	Ser	Gly	Asp	Asp	Ser	Leu	Leu	Asp	Arg	Glu	Pro	Pro	Thr	Arg
1775					1780						1785			
Pro	Glu	Trp	Val	Ile	Leu	Gln	Pro	Leu	Leu	Ser	Leu	Arg	Phe	Lys
1790					1795						1800			
Lys	Glu	Arg	Gly	Arg	Tyr	Ala	Thr	Phe	Cys	Gly	Tyr	Tyr	Ala	Ser

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His Val Gly Cys Val Arg Ser Pro Val Ala Leu Phe Ala Lys Leu		
1820	1825	1830
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Tyr Leu Ser Glu Phe Ala Leu Gly His Ser Leu Gly Asp His Leu		
1850	1855	1860
Trp Glu Ala Leu Pro Leu Glu Ala Val Pro Phe Gln Ser Ala Cys		
1865	1870	1875
Phe Asp Phe Phe Cys Arg Arg Ala Pro Arg His Leu Lys Leu Ser		
1880	1885	1890
Leu Met Leu Gly Glu Val Pro Glu Ser Ile Ile Ala Arg Ile Gly		
1895	1900	1905
Ser Ser Leu Lys Trp Ala Ser His Ala Ile Tyr Thr Thr Leu Ser		
1910	1915	1920
Ser Ala Ala Arg Val Ala Ile Leu Arg Ser Ser Arg Asn Ser Arg		
1925	1930	1935
Ser Met Pro Asp Asp Pro Asp Thr Thr Leu Leu Gln Gly Glu Leu		
1940	1945	1950
Leu Gln His Phe Gln Val Pro Phe Met Gln Ser Asp Thr Leu Leu		
1955	1960	1965
Pro Leu Thr Gly Gly Ser Ser Ala Pro Ile Leu Thr Pro Glu Ala		
1970	1975	1980
Phe Ser Thr Ser Leu Ala Phe Ser Met Ala Ser Asp Ala Gln Ala		
1985	1990	1995
Gly Pro Ala Pro Ser Arg Asp Asp Arg Val Asp Arg Gln Pro Arg		
2000	2005	2010
Leu Pro Ala Ala Pro Arg Val Ala Glu Val Gly Leu Asn Ala Pro		
2015	2020	2025
Ser Val Asp Tyr Pro Phe Gln Trp Val Val Ala Ser Tyr Asp Gly		
2030	2035	2040
Ser Glu Ala Lys Asn Leu Ser Asp Asp Leu Ser Gly Ser Ala Thr		
2045	2050	2055
Leu Thr Lys Val Met Ala Asn Tyr Arg His Ala Glu Leu Thr Ser		
2060	2065	2070
Val Glu Leu Glu Val Cys Pro Leu Ala Ala Phe Ser Lys Pro		
2075	2080	2085
Ile Ser Val Ser Ala Val Trp Thr Ile Ala Ser Ile Ser Pro Ala		
2090	2095	2100
Ser Ala Ser Glu Thr Ser Tyr Tyr Gly Gly Arg Leu Phe Thr Val		
2105	2110	2115
Gly Gly Pro Val Leu Met Ser Ser Thr Thr His Leu Pro Ala Asp		
2120	2125	2130
Leu Thr Arg Leu Asn Pro Val Leu Lys Gly Pro Val Lys Tyr Thr		
2135	2140	2145
Asp Cys Pro Arg Phe Ser Tyr Ser Val Tyr Ser Asn Gly Gly Thr		
2150	2155	2160
Lys Gly Thr Asn Leu Cys Thr Ile Ile Leu Arg Gly Val Val Arg		
2165	2170	2175
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2180	2185	

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<211> LENGTH: 154  
<212> TYPE: PRT  
<213> ORGANISM: Citrus Sudden Death Virus

<400> SEQUENCE: 3

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20          25          30

Gln Pro Ser Pro Ser Pro Ser Leu Cys Arg Pro Ser Gly Pro Leu Pro
35          40          45

Pro Ser Leu Gln Leu Pro Pro Leu Lys Pro Pro Thr Met Ala Val Asp
50          55          60

Ser Ser Leu Leu Ala Val Leu Ser Ser Cys Pro Ala Pro Pro Ile Ser
65          70          75          80

Leu Leu Ile Ser Pro Ala Ser Ile Leu Cys Ser Arg Ala Pro Ser Ser
85          90          95

Thr Gln Thr Ala Pro Asp Ser Pro Thr Pro Ser Thr Pro Met Ala Glu
100         105         110

Pro Arg Ala Pro Ile Ser Ala Pro Ser Ser Ser Gly Glu Leu Ser Ala
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Ser Ala Ala Pro Pro Val Ile Phe Ser Leu Arg Arg Ala Ser Ser Gly
130         135         140

Glu Gly Lys His Leu Leu Val Ser Ala Arg
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<210> SEQ ID NO 4  
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<213> ORGANISM: Artificial Sequence  
<220> FEATURE:  
<223> OTHER INFORMATION: Description of Artificial Sequence: Synthetic primer

&lt;400&gt; SEQUENCE: 4

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<210> SEQ ID NO 5  
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<213> ORGANISM: Artificial Sequence  
<220> FEATURE:  
<223> OTHER INFORMATION: Description of Artificial Sequence: Synthetic primer

&lt;400&gt; SEQUENCE: 5

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<210> SEQ ID NO 7  
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<212> TYPE: DNA

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<213> ORGANISM: Artificial Sequence  
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<223> OTHER INFORMATION: Description of Artificial Sequence: Synthetic primer

<400> SEQUENCE: 7

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24

<210> SEQ ID NO 8  
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<213> ORGANISM: Artificial Sequence  
<220> FEATURE:  
<223> OTHER INFORMATION: Description of Artificial Sequence: Synthetic peptide

<400> SEQUENCE: 8

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<210> SEQ ID NO 9  
<211> LENGTH: 14  
<212> TYPE: PRT  
<213> ORGANISM: Artificial Sequence  
<220> FEATURE:  
<223> OTHER INFORMATION: Description of Artificial Sequence: Synthetic peptide

<400> SEQUENCE: 9

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<210> SEQ ID NO 10  
<211> LENGTH: 14  
<212> TYPE: PRT  
<213> ORGANISM: Artificial Sequence  
<220> FEATURE:  
<223> OTHER INFORMATION: Description of Artificial Sequence: Synthetic peptide

<400> SEQUENCE: 10

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<210> SEQ ID NO 11  
<211> LENGTH: 14  
<212> TYPE: PRT  
<213> ORGANISM: Artificial Sequence  
<220> FEATURE:  
<223> OTHER INFORMATION: Description of Artificial Sequence: Synthetic peptide

<400> SEQUENCE: 11

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<210> SEQ ID NO 12  
<211> LENGTH: 15  
<212> TYPE: PRT  
<213> ORGANISM: Artificial Sequence  
<220> FEATURE:  
<223> OTHER INFORMATION: Description of Artificial Sequence: Synthetic peptide

<400> SEQUENCE: 12

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<210> SEQ ID NO 13  
<211> LENGTH: 6509  
<212> TYPE: DNA  
<213> ORGANISM: Oat Blue Dwarf Virus

<400> SEQUENCE: 13

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cgcaactgacc gccagccttc tcttcctttt gtcctctgtt ttgtggagag ctctctcgcc	5820
gtgccgcacg tcgacgtccc gttccaatgg gccgtcgctg cgtacgcgg agactccgc	5880
aagtttctca cccgacgaccc ctcaggatcc ttcacactga gccgcctcac catcggttat	5940
ccgcacgcgc agctcatctc cggcgatctc gagttggccc cccttgcgcg cgcctcgcc	6000
aagcccatct cccgttccgc cgtctggacc atagcctcca tggcccccgc caccaccacc	6060
gagctccagt actacgggtt ccgactcttc accctcgag gccccgtctt catgggttcc	6120
gtcacccgca tccctccgcg ctcacccgc ctcaaccccg tcatcaagac cggcggtggc	6180
ttcaactgact gccccggctt cacctactcc gtctatgcac acggcggttc cggcaacact	6240
cctctcatca cccgttcatgtt gcgaggagtt atccgccttc cggcccttc gggcaacacc	6300
gtcacccgcca cctaaaggccct ctcacccgtt tcaacaggag ttcttccttc gttttctcc	6360
tgacgaccaa tgaacgttgc ttatcccccc ttcaacatccc tccgtttccc cctccgtttt	6420
cctctctgtt ccattcccccc tctccctcc cgttcatcgca atgagtaagg ttccaggatcg	6480
attcaaagac ctgtatggat ttccctcg	6509

&lt;210&gt; SEQ\_ID NO 14

&lt;211&gt; LENGTH: 2066

&lt;212&gt; TYPE: PRT

&lt;213&gt; ORGANISM: Oat Blue Dwarf Virus

&lt;400&gt; SEQUENCE: 14

Met	Thr	Thr	Tyr	Ala	Phe	His	Pro	Leu	Leu	Pro	Thr	Pro	Thr	Ser	Phe
1				5			10				15				

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Ala Thr Ile Thr Gly Gly Leu Lys Asp Val Ile Glu Thr Leu Ser  
20 25 30

Ser Thr Ile His Arg Asp Thr Ile Ala Ala Pro Leu Met Glu Thr Leu  
35 40 45

Ala Ser Pro Tyr Arg Asp Ser Leu Arg Asp Phe Pro Trp Ala Val Pro  
50 55 60

Ala Ser Ala Leu Pro Phe Leu Gln Glu Cys Gly Ile Thr Val Ala Gly  
65 70 75 80

His Gly Phe Lys Ala His Pro His Pro Val His Lys Thr Ile Glu Thr  
85 90 95

His Leu Leu His Lys Val Trp Pro His Tyr Ala Gln Val Pro Ser Ser  
100 105 110

Val Leu Phe Met Lys Pro Ser Lys Phe Ala Lys Leu Gln Arg Gly Asn  
115 120 125

Ala Asn Phe Ser Ala Leu His Asn Tyr Arg Leu Thr Ala Lys Asp Thr  
130 135 140

Pro Arg Tyr Pro Asn Thr Ser Thr Ser Leu Pro Asp Thr Glu Thr Ala  
145 150 155 160

Phe Met His Asp Ala Leu Met Tyr Tyr Thr Pro Ala Gln Ile Val Asp  
165 170 175

Leu Phe Leu Ser Cys Pro Lys Leu Glu Lys Leu Tyr Ala Ser Leu Val  
180 185 190

Val Pro Pro Glu Ser Ser Phe Thr Ser Ile Ser Leu His Pro Asp Leu  
195 200 205

Tyr Arg Phe Arg Phe Asp Gly Asp Arg Leu Ile Tyr Glu Leu Glu Gly  
210 215 220

Asn Pro Ala His Asn Tyr Thr Gln Pro Arg Ser Ala Leu Asp Trp Leu  
225 230 235 240

Arg Thr Thr Thr Ile Arg Gly Pro Gly Val Ser Leu Thr Val Ser Arg  
245 250 255

Leu Asp Ser Trp Gly Pro Cys His Ser Leu Leu Ile Gln Arg Gly Ile  
260 265 270

Pro Pro Met His Ala Glu His Asp Ser Ile Ser Phe Arg Gly Pro Arg  
275 280 285

Ala Val Ala Ile Pro Glu Pro Ser Ser Leu His Gln Asp Leu Arg His  
290 295 300

Arg Leu Val Pro Glu Asp Val Tyr Asn Ala Leu Phe Leu Tyr Val Arg  
305 310 315 320

Ala Val Arg Thr Leu Arg Val Thr Asp Pro Ala Gly Phe Val Arg Thr  
325 330 335

Gln Cys Ser Lys Pro Glu Tyr Ala Trp Val Thr Ser Ser Ala Trp Asp  
340 345 350

Asn Leu Ala His Phe Ala Leu Leu Thr Ala Pro His Arg Pro Arg Thr  
355 360 365

Ser Phe Tyr Leu Phe Ser Ser Thr Phe Gln Arg Leu Glu His Trp Val  
370 375 380

Arg His His Thr Phe Leu Leu Ala Gly Leu Thr Thr Ala Phe Ala Leu  
385 390 395 400

Pro Pro Ser Ala Trp Leu Ala Asn Leu Val Ala Arg Ala Ser Ala Ser  
405 410 415

His Ile Gln Gly Leu Ala Leu Ala Arg Arg Trp Leu Ile Thr Pro Pro  
420 425 430

His Leu Phe Arg Pro Pro Pro Pro Ser Phe Ala Leu Leu Gln

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435	440	445	
Arg Asn Ser Thr Gly Pro Val Leu Leu Arg Gly Ser Arg		Glu Phe	
450	455	460	
Glu Ala Phe Pro Ser Leu Ala Pro Gln Leu Ala Arg Arg	Phe		
465	470	475	480
Leu Ala Arg Leu Leu Pro Gln Lys Pro Ile Asp Pro Trp	Val Val Ala		
485	490	495	
Ser Leu Ala Val Ala Val Ala Ile Pro Ala Ala Ser Leu	Ala Val Arg		
500	505	510	
Trp Phe Phe Gly Pro Asp Thr Pro Gln Ala Met His Asp	Arg Tyr His		
515	520	525	
Thr Met Phe His Pro Arg Glu Trp Arg Leu Thr Leu Pro	Arg Gly Pro		
530	535	540	
Ile Ser Cys Gly Arg Ser Ser Phe Ser Pro Leu Pro His	Pro Pro Ser		
545	550	555	560
Pro Thr Pro Ala Pro Asp Ser Arg Ala Glu Pro Leu Gln	Pro Pro Ser		
565	570	575	
Ala Pro Pro Ser Thr His Glu Pro Ala Pro Ala Asp Leu	Glu Pro Gln		
580	585	590	
Ala Pro Pro Ala His Ala Pro Gln Thr Glu Pro Pro Ser	Pro Val Ile		
595	600	605	
Glu Gln Glu Ala Arg Pro Asn Pro Leu Pro Ala Pro Ala	Pro Leu Ser		
610	615	620	
Ala Pro Thr Pro Ser Ala Ser Ala Pro Ser Leu Ala Pro	Thr Pro Ser		
625	630	635	640
Ala Pro Glu Pro Pro Ser Pro Thr Ala Ser Glu Gln Ala	Ala Ser Leu		
645	650	655	
Ile Pro Ala Pro Ser Ser Ala Leu Val Val Glu Pro Ser	Gly Val Val		
660	665	670	
Ser Ala Ser Ser Trp Gly Ala Thr Asn Gln Pro Ala Asp	Gln Val Asp		
675	680	685	
Asp Ser Pro Leu Ala Arg Asp Pro Ser Ala Ser Gly Pro	Val Arg Phe		
690	695	700	
Tyr Arg Asp Leu Phe Pro Ala Asn Tyr Ala Gly Asp Ser	Gly Thr Phe		
705	710	715	720
Asp Phe Arg Ala Arg Ala Ser Gly Arg Ser Pro Thr Pro	Tyr Pro Ala		
725	730	735	
Met Asp Cys Leu Leu Val Ala Thr Glu Gln Ala Thr Arg	Ile Ser Arg		
740	745	750	
Glu Ala Leu Trp Asp Cys Leu Thr Ala Thr Cys Pro Asp	Ser Phe Leu		
755	760	765	
Asp Pro Lys Ser Ile Ala Gln His Gly Leu Ser Thr Asp	His Phe Val		
770	775	780	
Ile Leu Ala His Arg Phe Ser Leu Cys Ala Asn Phe His	Ser Ala Glu		
785	790	795	800
His Val Ile Gln Leu Gly Met Ala Asp Ala Thr Ser Ile	Phe Met Ile		
805	810	815	
Asn His Thr Ala Gly Ser Ala Gly Leu Pro Gly His Phe	Ser Leu Arg		
820	825	830	
Leu Gly Asp Gln Pro Arg Ala Leu Asn Gly Gly Leu Ala	Gln Asp Leu		
835	840	845	
Ala Val Ala Ala Leu Arg Phe Asn Ile Ser Gly Asp Leu	Leu Pro Thr		
850	855	860	

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Arg Ser Val His Thr Tyr Arg Ser Trp Pro Lys Arg Ala Lys Asn Leu  
 865 870 875 880  
 Val Ser Asn Met Lys Asn Gly Phe Asp Gly Val Met Ala Ser Ile Asn  
 885 890 895  
 Pro Ile Arg Pro Ser Asp Ala Arg Glu Lys Ile Val Ala Leu Asp Gly  
 900 905 910  
 Leu Leu Asp Ile Ala Arg Pro Arg Ser Val Arg Leu Ile His Ile Ala  
 915 920 925  
 Gly Phe Pro Gly Cys Gly Lys Thr His Pro Ile Thr Lys Leu Leu His  
 930 935 940  
 Thr Ala Ala Phe Arg Asp Phe Lys Leu Ala Val Pro Thr Thr Glu Leu  
 945 950 955 960  
 Arg Ser Glu Trp Lys Glu Leu Met Lys Leu Ser Pro Ser Gln Ala Trp  
 965 970 975  
 Arg Phe Gly Thr Trp Glu Ser Ser Leu Leu Lys Ser Ala Arg Ile Leu  
 980 985 990  
 Val Ile Asp Glu Ile Tyr Lys Leu Pro Arg Gly Tyr Leu Asp Leu Ala  
 995 1000 1005  
 Ile His Ser Asp Ser Ser Ile Glu Phe Val Ile Ala Leu Gly Asp  
 1010 1015 1020  
 Pro Leu Gln Gly Glu Tyr His Ser Thr His Pro Ser Ser Ser Asn  
 1025 1030 1035  
 Ser Arg Leu Ile Pro Glu Val Ser His Leu Ala Pro Tyr Leu Asp  
 1040 1045 1050  
 Tyr Tyr Cys Leu Trp Ser Tyr Arg Val Pro Gln Asp Val Ala Ala  
 1055 1060 1065  
 Phe Phe Gln Val Gln Ser His Asn Pro Ala Leu Gly Phe Ala Arg  
 1070 1075 1080  
 Leu Ser Lys Gln Phe Pro Thr Thr Gly Arg Val Leu Thr Asn Ser  
 1085 1090 1095  
 Gln Asn Ser Met Leu Thr Met Thr Gln Cys Gly Tyr Ser Ala Val  
 1100 1105 1110  
 Thr Ile Ala Ser Ser Gln Gly Ser Thr Tyr Ser Gly Ala Thr His  
 1115 1120 1125  
 Ile His Leu Asp Arg Asn Ser Ser Leu Leu Ser Pro Ser Asn Ser  
 1130 1135 1140  
 Leu Val Ala Leu Thr Arg Ser Arg Thr Gly Val Phe Phe Ser Gly  
 1145 1150 1155  
 Asp Pro Ala Leu Leu Asn Gly Gly Pro Asn Ser Asn Leu Met Phe  
 1160 1165 1170  
 Ser Ala Phe Phe Gln Gly Lys Ser Arg His Ile Arg Ala Trp Phe  
 1175 1180 1185  
 Pro Thr Leu Phe Pro Thr Ala Thr Leu Leu Phe Ser Pro Leu Arg  
 1190 1195 1200  
 Gln Arg His Asn Arg Leu Thr Gly Ala Leu Ala Pro Ala Gln Pro  
 1205 1210 1215  
 Ser His Leu Leu Leu Pro Asp Leu Pro Ser Leu Pro Pro Leu Pro  
 1220 1225 1230  
 Ala Ser Gly Pro Tyr Ser Arg Ser Phe Pro Val Arg Ser Arg Phe  
 1235 1240 1245  
 Ala Ala Ala Val Lys Pro Ser Asp Arg Ser Asp Val Leu Ser Trp  
 1250 1255 1260

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Ala	Pro	Ile	Ala	Val	Gly	Asp	Gly	Glu	Thr	Asn	Ala	Pro	Arg	Ile
1265				1270							1275			
Asp	Thr	Ser	Phe	Leu	Pro	Glu	Thr	Arg	Arg	Pro	Leu	His	Phe	Asp
1280				1285							1290			
Leu	Pro	Ser	Phe	Arg	Pro	Gln	Ala	Pro	Pro	Pro	Ser	Asp	Pro	
1295					1300						1305			
Ala	Pro	Ser	Gly	Thr	Ala	Phe	Glu	Pro	Val	Tyr	Pro	Gly	Glu	Thr
1310					1315						1320			
Phe	Glu	Asn	Leu	Val	Ala	His	Phe	Leu	Pro	Ala	His	Asp	Pro	Thr
1325					1330						1335			
Asp	Arg	Glu	Ile	His	Trp	Arg	Arg	Gln	Leu	Ser	Asn	Gln	Phe	Pro
1340					1345						1350			
His	Val	Asp	Lys	Glu	Tyr	His	Leu	Ala	Ala	Gln	Pro	Met	Thr	Leu
1355					1360						1365			
Leu	Ala	Pro	Ile	His	Asp	Ser	Lys	His	Asp	Pro	Thr	Leu	Leu	Ala
1370					1375						1380			
Ala	Ser	Ile	Gln	Lys	Arg	Leu	Arg	Phe	Arg	Pro	Ser	Ala	Ser	Pro
1385					1390						1395			
Tyr	Arg	Ile	Ser	Pro	Arg	Asp	Glu	Leu	Leu	Gly	Gln	Leu	Leu	Tyr
1400					1405						1410			
Glu	Ser	Leu	Cys	Arg	Ala	Tyr	His	Arg	Ser	Pro	Thr	Thr	Thr	His
1415					1420						1425			
Pro	Phe	Asp	Glu	Ala	Leu	Phe	Val	Glu	Cys	Ile	Asp	Leu	Asn	Glu
1430					1435						1440			
Phe	Ala	Gln	Leu	Thr	Ser	Lys	Thr	Gln	Ala	Val	Ile	Met	Gly	Asn
1445					1450						1455			
Ala	Arg	Arg	Ser	Asp	Pro	Asp	Trp	Arg	Trp	Ser	Ala	Val	Arg	Ile
1460					1465						1470			
Phe	Ser	Lys	Thr	Gln	His	Lys	Val	Asn	Glu	Gly	Ser	Ile	Phe	Gly
1475					1480						1485			
Ala	Trp	Lys	Ala	Cys	Gln	Thr	Leu	Ala	Leu	Met	His	Asp	Ala	Val
1490					1495						1500			
Val	Leu	Leu	Leu	Gly	Pro	Val	Lys	Lys	Tyr	Gln	Arg	Val	Phe	Asp
1505					1510						1515			
Ala	Arg	Asp	Arg	Pro	Ala	His	Leu	Tyr	Ile	His	Ala	Gly	Gln	Thr
1520					1525						1530			
Pro	Ser	Ser	Met	Ser	Leu	Trp	Cys	Gln	Thr	His	Leu	Thr	Pro	Ala
1535					1540						1545			
Val	Lys	Leu	Ala	Asn	Asp	Tyr	Thr	Ala	Phe	Asp	Gln	Ser	Gln	His
1550					1555						1560			
Gly	Glu	Ala	Val	Val	Leu	Glu	Arg	Lys	Lys	Met	Glu	Arg	Leu	Ser
1565					1570						1575			
Ile	Pro	Asp	His	Leu	Ile	Ser	Leu	His	Val	His	Leu	Lys	Thr	His
1580					1585						1590			
Val	Glu	Thr	Gln	Phe	Gly	Pro	Leu	Thr	Cys	Met	Arg	Leu	Thr	Gly
1595					1600						1605			
Glu	Pro	Gly	Thr	Tyr	Asp	Asp	Asn	Thr	Asp	Tyr	Asn	Leu	Ala	Val
1610					1615						1620			
Ile	Asn	Leu	Glu	Tyr	Ala	Ala	Ala	His	Val	Pro	Thr	Met	Val	Ser
1625					1630						1635			
Gly	Asp	Asp	Ser	Leu	Leu	Asp	Phe	Glu	Pro	Pro	Arg	Arg	Pro	Glu
1640					1645						1650			
Trp	Val	Ala	Ile	Glu	Pro	Leu	Leu	Ala	Leu	Arg	Phe	Lys	Lys	Glu

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1655	1660	1665
Arg Gly	Leu Tyr Ala Thr Phe Cys	Gly Tyr Tyr Ala Ser Arg Val
1670	1675	1680
Gly Cys	Val Arg Ser Pro Ile Ala Leu Phe Ala Lys	Leu Ala Ile
1685	1690	1695
Ala Val	Asp Asp Ser Ser Ile Ser Asp Lys Leu Ala	Ala Tyr Leu
1700	1705	1710
Met Glu	Phe Ala Val Gly His Ser Leu Gly Asp Ser	Leu Trp Ser
1715	1720	1725
Ala Leu	Pro Leu Ser Ala Val Pro Phe Gln Ser Ala	Cys Phe Asp
1730	1735	1740
Phe Phe	Cys Arg Arg Ala Pro Arg Asp Leu Lys Leu	Ala Leu His
1745	1750	1755
Leu Gly	Glu Val Pro Glu Thr Ile Ile Gln Arg Leu	Ser His Leu
1760	1765	1770
Ser Trp	Leu Ser His Ala Val Tyr Ser Leu Leu Pro	Ser Arg Leu
1775	1780	1785
Arg Leu	Ala Ile Leu His Ser Ser Arg Gln His Arg	Ser Leu Pro
1790	1795	1800
Glu Asp	Pro Ala Val Ser Ser Leu Gln Gly Glu Leu	Leu Gln Thr
1805	1810	1815
Phe His	Ala Pro Met Pro Ser Leu Pro Ser Leu Pro	Leu Phe Gly
1820	1825	1830
Gly Leu	Ser Pro Asp Asn Ile Leu Thr Pro His Glu	Phe Arg Thr
1835	1840	1845
Ala Leu	Tyr Glu Ser Ser Ala Tyr Pro Thr Pro Pro	Asn Ser Pro
1850	1855	1860
Thr Ser	Met Ser Gly Ile His Ala Ser Gln Val Gly	Pro Pro Pro
1865	1870	1875
Ala Ser	Asp Asp Arg Thr Asp Arg Gln Pro Ser Leu	Pro Leu Ala
1880	1885	1890
Pro Arg	Ile Val Glu Ser Ser Leu Ala Val Pro His	Val Asp Val
1895	1900	1905
Pro Phe	Gln Trp Ala Val Ala Ser Tyr Ala Gly Asp	Ser Ala Lys
1910	1915	1920
Phe Leu	Thr Asp Asp Leu Ser Gly Ser Ser His Leu	Ser Arg Leu
1925	1930	1935
Thr Ile	Gly Tyr Arg His Ala Glu Leu Ile Ser Ala	Glu Leu Glu
1940	1945	1950
Phe Ala	Pro Leu Ala Ala Ala Phe Ala Lys Pro Ile	Ser Val Thr
1955	1960	1965
Ala Val	Trp Thr Ile Ala Ser Ile Ala Pro Ala Thr	Thr Thr Glu
1970	1975	1980
Leu Gln	Tyr Tyr Gly Gly Arg Leu Leu Thr Leu Gly	Gly Pro Val
1985	1990	1995
Leu Met	Gly Ser Val Thr Arg Ile Pro Ala Asp Leu	Thr Arg Leu
2000	2005	2010
Asn Pro	Val Ile Lys Thr Ala Val Gly Phe Thr Asp	Cys Pro Arg
2015	2020	2025
Phe Thr	Tyr Ser Val Tyr Ala Asn Gly Gly Ser Ala	Asn Thr Pro
2030	2035	2040
Leu Ile	Thr Val Met Val Arg Gly Val Ile Arg Leu	Ser Gly Pro
2045	2050	2055

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Ser Gly Asn Thr Val Thr Ala Thr  
2060 2065

<210> SEQ ID NO 15  
<211> LENGTH: 216  
<212> TYPE: PRT  
<213> ORGANISM: Grapevine Asteroid Mosaic-Associated Virus

&lt;400&gt; SEQUENCE: 15

Ser Ser Ala Pro Gln Leu Thr Ser Glu Ala Phe Ser Leu Thr Leu Ala  
1 5 10 15

Gln Ser Met Ala Ser Pro Asn Val Gln Ala Gly Pro Pro Pro Pro Ser  
20 25 30

Asp Asp Arg Thr Asp Arg Gln Pro Pro Leu Pro Arg Ala Pro Arg Leu  
35 40 45

Val Glu Asp Ala Ser Ala Ile Pro Phe Val Asp Tyr Pro Phe Gln Trp  
50 55 60

Val Val Ala Ser Tyr Asp Gly Ser Thr Ala Lys Asn Leu Thr Asp Val  
65 70 75 80

Leu Ser Gly Ser Lys Thr Leu Ser Thr Ile Thr Ala Asn Tyr Arg His  
85 90 95

Ala Glu Leu Leu Ser Val Glu Leu Glu Phe Ala Pro Leu Ala Gly Ser  
100 105 110

Phe Ser Lys Pro Ile Thr Leu Ser Ala Val Trp Thr Val Gly Ser Ile  
115 120 125

Thr Pro Ala Thr Thr Glu Thr Ser Tyr Tyr Gly Gly Arg Val Ile  
130 135 140

Thr Ile Gly Gly Pro Val Leu Met Asn Ser Thr Thr Ala Val Pro Ala  
145 150 155 160

Asp Leu Arg Arg Leu Asn Pro Ile Ile Lys Asp Gln Ile Ser Tyr Thr  
165 170 175

Asp Cys Pro Arg Phe Ser Tyr Ser Val Tyr Ala Asn Gly Gly Thr Ala  
180 185 190

Gly Thr Asn Leu Val Thr Val Leu Ile Arg Gly Val Val Arg Leu Arg  
195 200 205

Ser Pro Ser Gly Asn Leu Leu Ala  
210 215

<210> SEQ ID NO 16  
<211> LENGTH: 215  
<212> TYPE: PRT  
<213> ORGANISM: Citrus Sudden Death Virus

&lt;400&gt; SEQUENCE: 16

Ser Ser Ala Pro Ile Leu Thr Pro Glu Ala Phe Ser Thr Ser Leu Ala  
1 5 10 15

Phe Ser Met Ala Ser Asp Ala Gln Ala Gly Pro Ala Pro Ser Arg Asp  
20 25 30

Asp Arg Val Asp Arg Gln Pro Arg Leu Pro Ala Ala Pro Arg Val Ala  
35 40 45

Glu Val Gly Leu Asn Ala Pro Ser Val Asp Tyr Pro Phe Gln Trp Val  
50 55 60

Val Ala Ser Tyr Asp Gly Ser Glu Ala Lys Asn Leu Ser Asp Asp Leu  
65 70 75 80

Ser Gly Ser Ala Thr Leu Thr Lys Val Met Ala Asn Tyr Arg His Ala  
85 90 95

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Glu Leu Thr Ser Val Glu Leu Glu Val Cys Pro Leu Ala Ala Ala Phe  
 100 105 110

Ser Lys Pro Ile Ser Val Ser Ala Val Trp Thr Ile Ala Ser Ile Ser  
 115 120 125

Pro Ala Ser Ala Ser Glu Thr Ser Tyr Tyr Gly Gly Arg Leu Phe Thr  
 130 135 140

Val Gly Gly Pro Val Leu Met Ser Ser Thr Thr His Leu Pro Ala Asp  
 145 150 155 160

Leu Thr Arg Leu Asn Pro Val Leu Lys Gly Pro Val Lys Tyr Thr Asp  
 165 170 175

Cys Pro Arg Phe Ser Tyr Ser Val Tyr Ser Asn Gly Gly Thr Lys Gly  
 180 185 190

Thr Asn Leu Cys Thr Ile Ile Leu Arg Gly Val Val Arg Leu Ser Gly  
 195 200 205

Pro Ser Gly Asn Leu Leu Ala  
 210 215

<210> SEQ ID NO 17  
<211> LENGTH: 198  
<212> TYPE: PRT  
<213> ORGANISM: Grapevine Asteroid Mosaic-Associated Virus

&lt;400&gt; SEQUENCE: 17

Met Ala Ser Pro Asn Val Gln Ala Gly Pro Pro Pro Pro Ser Asp Asp  
 1 5 10 15

Arg Thr Asp Arg Gln Pro Pro Leu Pro Arg Ala Pro Arg Leu Val Glu  
 20 25 30

Asp Ala Ser Ala Ile Pro Phe Val Asp Tyr Pro Phe Gln Trp Val Val  
 35 40 45

Ala Ser Tyr Asp Gly Ser Thr Ala Lys Asn Leu Thr Asp Val Leu Ser  
 50 55 60

Gly Ser Lys Thr Leu Ser Thr Ile Thr Ala Asn Tyr Arg His Ala Glu  
 65 70 75 80

Leu Leu Ser Val Glu Leu Glu Phe Ala Pro Leu Ala Gly Ser Phe Ser  
 85 90 95

Lys Pro Ile Thr Leu Ser Ala Val Trp Thr Val Gly Ser Ile Thr Pro  
 100 105 110

Ala Thr Thr Thr Glu Thr Ser Tyr Tyr Gly Gly Arg Val Ile Thr Ile  
 115 120 125

Gly Gly Pro Val Leu Met Asn Ser Thr Ala Val Pro Ala Asp Leu  
 130 135 140

Arg Arg Leu Asn Pro Ile Ile Lys Asp Gln Ile Ser Tyr Thr Asp Cys  
 145 150 155 160

Pro Arg Phe Ser Tyr Ser Val Tyr Ala Asn Gly Gly Thr Ala Gly Thr  
 165 170 175

Asn Leu Val Thr Val Leu Ile Arg Gly Val Val Arg Leu Arg Ser Pro  
 180 185 190

Ser Gly Asn Leu Leu Ala  
 195

<210> SEQ ID NO 18  
<211> LENGTH: 197  
<212> TYPE: PRT  
<213> ORGANISM: Citrus Sudden Death Virus

&lt;400&gt; SEQUENCE: 18

-continued

Met Ala Ser Asp Ala Gln Ala Gly Pro Ala Pro Ser Arg Asp Asp Arg  
 1 5 10 15  
 Val Asp Arg Gln Pro Arg Leu Pro Ala Ala Pro Arg Val Ala Glu Val  
 20 25 30  
 Gly Leu Asn Ala Pro Ser Val Asp Tyr Pro Phe Gln Trp Val Val Ala  
 35 40 45  
 Ser Tyr Asp Gly Ser Glu Ala Lys Asn Leu Ser Asp Asp Leu Ser Gly  
 50 55 60  
 Ser Ala Thr Leu Thr Lys Val Met Ala Asn Tyr Arg His Ala Glu Leu  
 65 70 75 80  
 Thr Ser Val Glu Leu Glu Val Cys Pro Leu Ala Ala Ala Phe Ser Lys  
 85 90 95  
 Pro Ile Ser Val Ser Ala Val Trp Thr Ile Ala Ser Ile Ser Pro Ala  
 100 105 110  
 Ser Ala Ser Glu Thr Ser Tyr Tyr Gly Gly Arg Leu Phe Thr Val Gly  
 115 120 125  
 Gly Pro Val Leu Met Ser Ser Thr Thr His Leu Pro Ala Asp Leu Thr  
 130 135 140  
 Arg Leu Asn Pro Val Leu Lys Gly Pro Val Lys Tyr Thr Asp Cys Pro  
 145 150 155 160  
 Arg Phe Ser Tyr Ser Val Tyr Ser Asn Gly Gly Thr Lys Gly Thr Asn  
 165 170 175  
 Leu Cys Thr Ile Ile Leu Arg Gly Val Val Arg Leu Ser Gly Pro Ser  
 180 185 190  
 Gly Asn Leu Leu Ala  
 195

<210> SEQ ID NO 19  
 <211> LENGTH: 309  
 <212> TYPE: PRT  
 <213> ORGANISM: Grapevine Fleck Virus

<400> SEQUENCE: 19

Met Thr Ser Arg Ala Pro Ser Pro Pro Thr Pro Pro Cys Pro Ser Pro  
 1 5 10 15  
 Pro Ala Leu Lys Ser Ser Pro Ser Pro Val Pro Thr Ala Thr Pro Ala  
 20 25 30  
 Ser Pro Pro Leu Lys Pro Leu Ser Asn Pro Leu Pro Pro Pro Pro  
 35 40 45  
 Thr Pro Arg Pro Ser Thr Ser Ala Gly Pro Ser Thr Pro Leu Pro Pro  
 50 55 60  
 Pro Ala Leu Arg Ser Ser Pro Ser Ser Ala Leu Asn Ala Ser Arg Gly  
 65 70 75 80  
 Ala Pro Ser Thr Ser Pro Pro Ser Ser Ser Pro Pro Ser Ser Pro  
 85 90 95  
 Ala Ser Thr Pro Pro Ser Arg Thr Pro Ser Pro Thr Pro Thr Ala Pro  
 100 105 110  
 Ala Ser Pro Val Ala Ser Thr Ala Met Thr Pro Ala Ser Pro Ser Val  
 115 120 125  
 Pro Pro Pro Pro Ser Ala Ala Pro Ser Ser Ser Ala Ala Leu Ser Ser  
 130 135 140  
 Ala Pro Pro Pro Ser Thr Ala Pro Leu Pro Arg His Glu Pro Arg Pro  
 145 150 155 160  
 Pro Pro Pro Leu Pro Pro Pro Leu Gln Pro Pro Pro Gly Val Arg Val

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165	170	175
Pro Arg Ser Val Ala Phe Pro Leu Pro Leu Ala Arg Glu Leu Pro Pro		
180	185	190
Leu Arg Leu Pro Pro Ala Pro Tyr Leu His Pro Leu Leu Ala Arg Leu		
195	200	205
Ala Pro Leu Arg Leu Arg Pro Pro Pro Asp Leu Pro Ser Pro Pro Leu		
210	215	220
Ser Pro Pro Leu Ser Pro Pro Leu Ser Pro Ile Ser Pro Leu His Ala		
225	230	235
Pro Ala Pro Pro His Pro Asp Pro Val Leu Leu Pro Ala Leu Ser		
245	250	255
Leu Ala Ile Ser Arg Ala Ala Pro Asp Leu Leu Arg Leu Leu Ser Leu		
260	265	270
Leu Ser Pro Pro Ser Leu Phe Leu Leu Phe Thr Leu Leu Ser Ile His		
275	280	285
Phe Ser Pro Phe Pro Ile Phe Ile Leu Leu Ser Leu Leu Leu Leu		
290	295	300
Gln Phe Pro Arg Thr		
305		

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The invention claimed is:

1. A recombinant expression vector comprising a cDNA molecule comprising a nucleotide sequence encoding an amino acid sequence having amino acids 1974-2188 of SEQ ID NO: 2, operably linked to a heterologous promoter.
2. An isolated nucleic acid molecule of claim 1, wherein said nucleotide sequence further encodes amino acids 1992-2188 of SEQ ID NO: 2.
3. A recombinant expression vector of claim 1, wherein said nucleotide sequence further encodes amino acids 127-337 of SEQ ID NO: 2.
4. A recombinant expression vector of claim 1, wherein said nucleotide sequence further encodes amino acids 897-1002 of SEQ ID NO: 2.

5. A recombinant expression vector of claim 1, wherein said nucleotide sequence further encodes amino acids 1084-1315 of SEQ ID NO: 2.

6. A recombinant expression vector of claim 1, wherein said nucleotide sequence further encodes amino acids 1-154 of SEQ ID NO: 3.

7. A recombinant expression vector of claim 1, wherein said nucleotide sequence further encodes amino acids 1474-1890 of SEQ ID NO: 2.

8. A cDNA molecule comprising a nucleotide sequence consisting of nucleotides 6028-6675 of SEQ ID NO: 1, operably linked to a heterologous promoter.

\* \* \* \* \*